Frequency-Dependent Gating of Hippocampal–Neocortical Interactions

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Abstract

How and where hippocampal–neocortical interactions required for memory formation take place is a major issue of current research. Using a combined in vivo functional magnetic resonance imaging/electrophysiology approach, we have investigated whether specific frequencies of CA3 neuronal activation, inducing different forms of short-term plasticity at CA1 synapses, contribute to differential activity propagation in brain-wide networks connected to the hippocampus. We report that localized activation of CA3 neurons in dorsal hippocampus produced activity propagation within the hippocampal formation, including the subiculum and entorhinal cortex, which increased monotonically with frequency to a maximum at 20–40 Hz. However, robust extrahippocampal propagation was seen specifically at theta–beta frequencies (10–20 Hz), reaching a network of midline neocortical and mesolimbic structures. Activation in those regions correlated with a frequency-dependent facilitation of spiking activity recorded in CA1. These results provide a mechanistic link between the dynamic properties of short-term plasticity in the efferent synapses of CA3 neurons in CA1 and activity propagation in brain-wide networks, and identify polysynaptic information channels segregated in the frequency domain.

Key words: activity propagation, beta oscillations, electrophysiology, fMRI, gating, hippocampus, memory, neocortex, rat, short-term plasticity, theta oscillations

Introduction

A recurrent theme in the study of memory is the need for interactions between the hippocampus and the neocortex when new information is incorporated (McNaughton and Morris 1987; Buzsaki 1989; Squire 1992; McClelland et al. 1995; Morris 2006; Girardeau et al. 2009). However, the spatiotemporal nature and causal consequences of these physiological interactions are not yet fully understood.

A number of studies using different techniques to map brain activity globally, such as detection of 2DG uptake (Bontempi et al. 1999) and immediate early genes (Maviel et al. 2004; Frankland and Bontempi 2005), have been used to identify distributed networks of structures recruited during specific behavioral tasks or stimulation paradigms. These have been valuable in identifying activation in a set of midline structures of the neocortex following hippocampal-dependent learning, but more dynamic accounts of connectivity that causally relates activity in the hippocampus to cortical regions are desirable. An alternative method is the use of functional magnetic resonance imaging (fMRI) with blood–oxygen level-dependent (BOLD) contrast combined with targeted activation of brain nuclei (i.e., by means of electrical microstimulation) and concomitant electrophysiological recordings (Tolias et al. 2005; Angenstein et al. 2007; Canals, Beyerlein, Murayama et al. 2008; Logothetis et al. 2010; Moreno et al. 2013; Alvarez-Salvado et al. 2014; Weitz et al. 2014). We have used this technique to investigate whether short-term plasticity of synapses, induced by distinct frequencies of CA3 neuronal activation and recorded in
CA1, conditions activity transfer from the hippocampus to the neocortex. Using this novel experimental approach, we found several functional connectivity patterns for CA3 depending on its frequency of activation. Overall, our findings point to a mechanistic link between the dynamic properties of hippocampal synapses and those of activity propagation in brain-wide neuronal networks.

Materials and Methods

Animals

Data from 13 male Sprague-Dawley rats (250–300 g) are reported from combined electrophysiology and fMRI studies; a further 5 rats were used in pilot work. Animals were purchased from Janvier Labs (France) and maintained under a 12/12-h light/dark cycle (lights on 07:00–19:00 h) at room temperature (22 ± 2°C), with free access to food and water. Rats were housed in groups of 5 and adapted to these conditions for at least 1 week before experimental manipulation. All experiments were approved by the local authorities (IN-CSIC) and were performed in accordance with Spanish (law 32/2007) and European regulations (EU directive 86/609, EU decree 2001-486).

Carbon-Fiber Electrodes

Glass-coated carbon-fiber bipolar electrodes were developed for the present study based on previous reports (Shyu et al. 2004). Individual 7 μm diameter carbon fibers (Goodfellow Cambridge Limited, England) were used. These consisted of bundles of fibers inserted into a theta-shaped glass capillary (World Precision Instruments) previously pulled to form 7 mm long pipettes with ~200 μm tip diameter and adjusted to produce an electrical impedance of 40–65 kΩ. A regular wire with a pin connector was attached to the pipette, connected to the carbon fibers using silver conductive epoxy resin (RS Components, UK), and isolated with clear epoxy resin. Afterwards, the tips was bent in a flame to form a 90° angle to minimize implant size, and thereby allow close proximity between the magnetic resonance imaging (MRI) array coil and the head of the animal. These electrodes were stereotaxically implanted to target specific locations in the dorsal hippocampus with a final arrangement as shown in Figure 1 and Supplementary Figure 1.

Electrode Implantation and Electrophysiology

All experiments were performed under urethane anesthesia (1.3 g/kg, i.p.). Stimulating and recording electrodes were implanted using standard surgical and stereotaxic procedures, as described previously (Canals, Beyerlein, Keller et al. 2008; Canals, Beyerlein, Murayama et al. 2008; Canals et al. 2009). A carbonfiber bipolar stimulating electrode was positioned in the CA3 region of the dorsal hippocampus (from bregma: 3.5 mm anteroposterior and 3.5 mm lateral, initial position 3.6 mm ventral to the dural surface). An electrophysiological multichannel recording electrode (single shank, 100 μm contact spacing, 32 channels; Neuronexus Technologies) was targeted to the contralateral CA1 (3.5 mm caudal and 2.5 mm lateral from bregma). The final position of the bipolar carbon electrode was adjusted to the stratum pyramidale according to electrophysiological potentials recorded in CA1 (Fig. 1C). Stimulation along the implantation tract of the carbon-fiber electrode, from dorsal stratum oriens to the ventral border of CA3 demonstrates 2 peaks of excitatory postsynaptic potentials (EPSPs) recorded in the contralateral CA1, the first evoked by the activation of Schaffer collateral/commissural axons in stratum radiatum and the second by the depolarization of CA3 neurons in the soma cell layer (Supplementary Fig. 2). The second one was selected for the present experiments.

The multichannel electrode was then substituted by an MRI-compatible L-shaped single channel borosilicate electrode to record the population spike (PS) in the contralateral CA1 (Supplementary Fig. 1). Stimulating and recording electrodes were secured to the skull with dental cement (Heraeus Medical, Beyerlein, Murayama et al. 2008; Canals, Beyerlein, Murayama et al. 2008; Canals et al. 2009). A carbon-fiber bipolar stimulating electrode was positioned in the CA3 region of the dorsal hippocampus (from bregma: 3.5 mm anteroposterior and 3.5 mm lateral, initial position 3.6 mm ventral to the dural surface). An electrophysiological multichannel recording electrode (single shank, 100 μm contact spacing, 32 channels; Neuronexus Technologies) was targeted to the contralateral CA1 (3.5 mm caudal and 2.5 mm lateral from bregma). The final position of the bipolar carbon electrode was adjusted to the stratum pyramidale according to electrophysiological potentials recorded in CA1 (Fig. 1C). Stimulation along the implantation tract of the carbon-fiber electrode, from dorsal stratum oriens to the ventral border of CA3 demonstrates 2 peaks of excitatory postsynaptic potentials (EPSPs) recorded in the contralateral CA1, the first evoked by the activation of Schaffer collateral/commissural axons in stratum radiatum and the second by the depolarization of CA3 neurons in the soma cell layer (Supplementary Fig. 2). The second one was selected for the present experiments.

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Figure 1. Experimental setup. (A) Micrograph of the tip of one of the carbon-fiber bipolar microstimulating electrodes used in the study. CF (carbon fiber), S (septum). (B) Coronal section of Paxinos and Watson atlas (Paxinos and Watson 2007) at 3.5 mm posterior to bregma illustrating the position of the microstimulating (asterisk) and recording (diamond) electrodes. (Inset) Schematic representation of the L-shaped electrodes implanted in the rat dorsal hippocampus. The vertical plane (dark gray) depicts the approximate location of the coronal section from the atlas and the MR image in (D). (C) Electrophysiological field potentials recorded along the dorsoventral extension of CA1 were used to optimize the effective positioning of the carbon-fiber microstimulation in the contralateral CA3 first, and then to quantify CA1 spiking activity during the experiment. Note the typical EPSP (arrow) and PS recorded in the stratum radiatum and pyramidale, respectively. (D) Representative T2-weighted MRI anatomy with overlaid functional maps evoked by CA3 stimulation. Note the very small artifact in the MR image produced by the carbon-fiber electrode (asterisk) and the virtual absence of artifacts induced by the borosilicate recording electrode (diamond). Scale bars: 100 μm (A), 2 ms, 4 mV (C), 5 mm (D).
Wehrheim, Germany) and the animal then transferred to the scanner. These arrangements ensured the specificity of the stimulation during fMRI acquisition and simultaneous electrophysiological recording.

After filtering (0.1 Hz–3 kHz) and amplification, the electrophysiological signals were digitized (20 kHz acquisition rate) and stored for offline processing with MatLab (The MathWorks, USA) and Spike2 (Cambridge Electronic Design, Cambridge, USA). The EPSP, reflecting the population synaptic response, was measured as the maximal slope of the negative going field potential recorded in CA1 stratum radiatum, and the PS in the CA1 pyramidal layer, reflecting the summed spiking activity of the recorded population, as the amplitude between the maximal negativity and the preceding positive crest. To correlate the fast electrophysiological responses and the slow BOLD signals for every stimulation protocol used (see below), we calculated the mean PS amplitude in response to all pulse in a stimulation train (equivalent to the area under the curves in Fig. 4D). Those integrated measurements of spiking activity were then correlated to the corresponding BOLD signal amplitudes (see below).

Stimulation Protocols

Charge balanced biphasic 0.1-ms duration pulses were delivered through the carbon-fiber stimulating electrode with a constant current source and a pulse generator (STG2004, Multichannel Systems, Reutlingen, Germany). Importantly, none of the stimulation protocols used induced episodes of spontaneous (epileptiform) activity (Supplementary Fig. 3).

For fMRI data acquisition, multiple stimulation protocols were used (Supplementary Fig. 4). Each protocol consisted of 10 trains of stimulation with trains repeated every 30 s (300 s per protocol) and protocols repeated 3 times per condition. In a first set of conditions, the trains were designed to maintain a constant charge transfer by fixing the total number of pulses to 40 and electric pulse width to 100 μs (biphasic), but varying the stimulation frequencies between 5, 10, 20, and 40 Hz. These protocols yielded stimulation trains of 8, 4, 2, and 1 s, respectively, and were presented in random order. In a second set of conditions, the different stimulation trains were mixed in a single protocol in pseudorandom order (Fig. 4A). In a third set of conditions, the protocols were designed with a constant duration of the stimulus train at 10 and 40 Hz (4 s), thereby delivering a different number of pulses (40 and 160, respectively); or in which the stimulation frequency was increased to 160 Hz (1 s, 160 pulses/train). A final set of conditions consisted of 40 Hz stimulation trains structure into theta- and delta-burst protocols. In these protocols, one stimulation period consisted of short trains of 4 and 8 pulses separated by off periods of 250 and 600 ms and repeated 10 and 5 times, respectively. Stimulation periods were repeated 10 times as before.

MRI Experiments and Data Analysis

For the MRI experiments, the previously prepared urethane-anesthetized animals were placed in a custom-made animal holder with adjustable bite and ear bars, and positioned on the magnet bed. The animals were constantly supplied with 0.8 L/m 37.5°C through a water heat-pad. The temperature, heart rate, FA, 60° (time echo) TE, 15 ms; (time repetition) TR, 2000 ms. T2-weighted anatomical images were collected using a rapid acquisition relaxation enhanced sequence (RARE): FOV, 25 × 25 mm; 15 slices; slice thickness, 1 mm; matrix, 192 × 192; TEeff, 56 ms; TR, 2 s; RARE factor, 8. A 1H rat brain receive-only phased-array coil with integrated combiner and preamplifier, and no tune/no match, was employed in combination with the actively detuned transmit-only resonator (Bruker BioSpin MRI GmbH, Germany).

Functional MRI data were analyzed offline using our own software developed in MatLab, which included Statistical Parametric Mapping package (SPM, www.fil.ion.ucl.ac.uk/spm), Analysis of Functional NeuroImages (AFNI, http://afni.nimh.nih.gov/afni) and FSL Software (FMRIB http://fsl.fmrib.ox.ac.uk/fsl/). After linear detrending, temporal (0.015–0.2 Hz) and spatial filtering (3 × 3 gaussian kernel of 1.5 sigma) of voxel time series, a general linear model (GLM) or cross-correlation analysis was applied with a simple boxcar model shifted forward in time, typically by 2 s, or a boxcar convolved with the hemodynamic response function (HRF) (MatLab). The results were largely comparable with all methods tested (GLM with HRF was used in Fig. 3 and cross-correlation analysis results with HRF are shown in Supplementary Fig. 5). Functional maps were generated from voxels that had a highly significant (P < 0.0001) component for the model and were clustered together in space (cluster size = 14; calculated with Monte Carlo simulation implemented in AFNI).

Regions of interest (ROIs) extracted using a rat atlas registered to the functional images (Schwarz et al. 2006) (Supplementary Fig. 6) were used to compute the amplitude of the evoked BOLD signal responses (as a percentage relative to a prestimulus baseline of 6 s) and volume of brain tissue activated in absolute terms (number of voxels above the statistical threshold) or relative to the ROI (number of active voxels divided by the total number of voxels in the region). The maps shown in Figure 3 represent the group probability for each voxel of being activated by a particular stimulation protocol. A voxel probability of 1 means that 9 out of 9 animals used in that study showed coincident activation of that particular voxels to a certain protocol. The resulting maps are color-coded and thresholded so that probabilities <0.33 (activations found in only 3 out of 9 animals) are not represented. Shown are 7 coronal slices from caudal (left, ~8 mm from bregma) to rostral (right, ~4 mm from bregma).

Histology

After completion of each experiment, the rats were perfused intracardially with 100 mL of 1% phosphate-buffered saline (PBS) solution and 50 mL of ice-cold 4% paraformaldehyde (PFA). Brains were kept for 24 h on 4% PFA post-fixation at 4°C and cut in a fixed material vibratome in 50 μm thick slices. Slices were then stained with 4',6-diamidino-2-phenylindole (DAPI) for photography under a fluorescence microscope. The position of the electrodes was confirmed by the observable damage of the insertion.

Results

We have combined fMRI and local field potential (LFP) recordings in the rat (Canals, Beyerlein, Murayama et al. 2008; Canals, Beyerlein et al. 2009) to investigate brain-wide patterns of activity propagation as a function of the frequency of neuronal activation
Activity Propagation in Brain-Wide Hippocampal Networks

We used fMRI stimulation protocols consisting of 10 repetitions of an ON-OFF sequence with a duty-cycle of 30 s, known to produce repetitive and reliably stable BOLD signals (Canals, Beyerlein, Murayama et al. 2008). Stimulation parameters during the ON-period were chosen according to the temporal characteristics of action potential activity of hippocampal neurons observed in the behaving animal (Ranck 1973; Berger et al. 1983). Stimulation in all experiments was set at a frequency equal to 5 Hz (Supplementary Fig. 1). We implanted these and MRI-compatible recording electrodes precisely guided by layer-specific potentials to target the stratum pyramidale of CA3 and CA1, respectively (Fig. 1B, Supplementary Fig. 2). Bipolar depolarizing pulses delivered through the CA3 carbon-fiber electrodes generated localized LFP activation within CA1 (Fig. 1C), but also more widespread fMRI signals that we used as readout of activity propagated from the CA3 network (Fig. 1D).

Short-Term Plasticity at CA3→CA1 Synapses and Activity Propagation

Frequency-dependent and short-term changes in synaptic transmission in the Schaffer collateral and spiking activity in CA1 have been previously described from low- to high-stimulation frequencies (Alger and Nicoll 1982; Herreras et al. 1987; Dutar and Nicoll 1988; Davies and Collingridge 1993; Davies and Collingridge 1996). Accordingly, we measured CA1 PS amplitude corresponding to the 4 stimulation protocols (5, 10, 20, and 40 Hz) recorded in our combined fMRI-electrophysiology experiments. We observed that spiking activity was largely facilitated in the range of propagating frequencies (10–20 Hz). However, comparable facilitation did not occur at 5 Hz and fell away at 40 Hz (Fig. 4D). We computed the averaged PS amplitude across all delivered pulses as an integral measure of the output spiking activity, for each animal at each tested frequency. The results of this analysis unveiled an inverted U-shape curve with maxima at 10–20 Hz and minima at 5 and 40 Hz (Fig. 4E), reminiscent of those found for the fMRI signals in extrahippocampal areas (Fig. 4C). Accordingly, we plotted fMRI magnitude as a function of PS amplitude and observed a linear correlation. This finding suggests a mechanistic link between local PS facilitation as a consequence of short-term plasticity and activity propagation to extrahippocampal targets (Fig. 4F).

Frequency Determines Hippocampal–neocortical Interactions

Further control studies were necessary as it should be noted that, in addition to alteration in frequency, the choice of 40 pulses per duty cycle meant that the duration of stimulation covaried. We therefore dissected the potential contributions of stimulus frequency versus duration. Train duration was kept constant to 4 s, with the stimulation frequency alternating between 10 and 40 Hz, thereby delivering 40 and 160 pulses, respectively (Supplementary Fig. 4, compare B,E). Extensive and indistinguishable activations in the hippocampal formation were found with both protocols (Fig. 5A), with neocortical propagation occurring at 10 Hz but not at 40 Hz (Fig. 5B), even though 4 times more pulses were delivered to CA3 in the second protocol. In a next set of experiments, even higher stimulation frequencies were tested by delivering 160 pulses in 1 s (160 Hz; Supplementary Fig. 4F), leading to the identical result as 40 Hz stimulation (Fig. 5). Given observations suggesting that spike amplitude would first climb and then decline at 40 Hz (i.e., Fig. 4D), we also examined high frequency (40 Hz) stimulation bursts of 4–8 pulses separated by 250 or 600 ms, respectively (theta-to-delta burst stimulation). Once again, we observed that this produced extensive hippocampal activation in the absence of extrahippocampal propagation, (Fig. 5, data for both burst protocols pooled in the same analysis for simplicity). Overall, these findings robustly indicate that the frequency of hippocampal activation is the critical factor.
determining the extent of propagation to cortical and subcortical structures.

**Discussion**

The main findings of this study are that (1) localized activation of a discrete area of CA3 neurons propagates across hippocampal subfields, including the Schaffer collateral/commissural output of CA3 to CA1, and may then invade spatially diverse neocortical and subcortical territories; (2) propagation inside the hippocampal formation increases monotonically with frequencies from 5 to 40 Hz, reaching a plateau at 20–40 Hz; (3) robust propagation beyond the hippocampal formation occurs specifically at frequencies between 10 and 20 Hz, independently of train duration; (4) extrahippocampal activity propagation in fMRI measurements correlates with the magnitude of spiking activity recorded...
in CA1, which is dynamically determined by short-term plasticity; (5) stimulation at 10 and 20 Hz induces frequency facilitation of CA1 PS output; and (6) the afferent neurons receiving propagated hippocampal activity configure a network of frontoparietal neocortical structures and mesolimbic dopaminergic regions.

Synaptic Physiology Level

Our results suggest that the output of CA3 implements a frequency-dependent gating mechanism that promotes long-range interactions at 10–20 Hz. The primary aim of our study was to delineate the characteristics of any hippocampal gating of propagation rather than identify mechanisms, but several possibilities can be envisioned based on the previous work. For example, we show that the spiking activity of CA1 pyramidal neurons is facilitated in a train at propagating frequencies, a finding in good agreement with extensive previous literature in hippocampal slices demonstrating frequency-facilitation of neural transmission in CA1 at theta–beta ranges (Alger and Nicoll 1982a, b; Dutar and Nicoll 1988; Davies and Collingridge 1993; Davies and Collingridge 1996). This facilitation has been shown to be the combined result of presynaptic GABA_A-mediated decrease in synaptic inhibition and the concomitant enhancement of glutamate release. Conversely, at higher frequencies (starting at 40–50 Hz), a GABA_A-mediated inhibitory current supported by recurrent interneuronal networks builds up progressively in the train strongly curtailting CA1 pyramidal cell firing (Pouille and Scanziani 2004). The dynamic nature of these opposing short-term plasticity processes is clearly illustrated in the behavior of the PS amplitude at 40 Hz (i.e., Fig. 4D, blue tracing) in which an initial facilitation is then efficiently curtailed after several pulses.

It is therefore likely that the range of frequency-dependent facilitation of activity propagation beyond hippocampus found in our study is determined by interplay of the biophysical properties of pre- and postsynaptic elements in the CA3–CA1 circuits, including principal cells and interneurons, and their short-term plasticity. While the failure of fMRI signals to propagate at high frequencies occurs simultaneously with maximal amplitude signals locally in the hippocampus, the failure at lower frequencies coincides with the lowest activation inside the hippocampus (Fig. 4). We hypothesize (see above) that the lack of spiking facilitation at low frequencies precludes the boosting of fMRI signals propagation, however, a contribution of limited sensitivity to detect BOLD signals at low frequencies cannot be fully discarded (Canals, Beyerlein, Murayama et al. 2008). Future electrophysiological recordings simultaneously in the hippocampus and those cortical regions targeted by activity propagation will try to clarify this extent focusing on ongoing activity propagation (Fernández-Ruiz et al. 2012). Also, a potential contribution of the septum, which receives monosynaptic inputs from CA3 neurons and is activated in our fMRI experiments, cannot be ruled out with the present experiments. In addition to the identified functional roles of the dynamic CA3–CA1 interaction such as the modulation of synaptic plasticity (Davies et al. 1991; Mott and Lewis 1991; Manabe et al. 1993; Perkel and Nicoll 1993) and...
temporal fidelity of pyramidal cell firing (Pouille and Scanziani 2001; Schaefer et al. 2006), our results suggest a new role of this local hippocampal circuit in routing activity propagation according to input frequency.

Synapse to Network Transformations

Activation of CA3 neurons at 10–20 Hz is routed towards a distributed but highly reproducible network of brain structures known to be critically related to memory formation, including the PFC. Interestingly, in previous fMRI studies we showed that long-term potentiation (LTP) of synaptic strength in the DG—the accepted experimental analog of learning at the synaptic level—induces a functional reorganization of the network such that test stimulation delivered at 10 Hz which produces activations initially circumscribed to the hippocampal formation, started to propagate to the PFC and ventral striatum (Canals, Beyerlein et al. 2009). Similarly, electrophysiological evidence from other groups further has shown that LTP induction in the same pathway facilitates the disynaptic transfer of activity at 10 Hz from the DG to CA3 (Yeckel and Berger 1998). In this context, the previously described functional reorganization of hippocampal outputs after

Figure 4. Short-term plasticity facilitates CA1 output and boosts system-level interactions. (A) fMRI BOLD signals evoked in the hippocampus (upper panel) and PFC (lower panel) by a pseudorandom presentation of stimulation frequencies. Inset indicates the time of presentation of the different protocols. Colors in all panels represent stimulation frequencies as follows: red (5 Hz), orange (10 Hz), green (20 Hz), and blue (40 Hz). Values represent mean ± SEM. (B) BOLD signal amplitude (% from baseline, n = 9, P < 0.0001, cluster size 14) in dorsal hippocampus and (C) PFC across stimulation frequencies. PFC is composed of frontal, orbital, prelimbic, infralimbic, cingulate, and insular cortices. Note the monotonic increase in hippocampal BOLD signals amplitude (repeated-measures one-way ANOVA F3,24 = 37.05, P < 0.0001) versus the inverted U-shape response for PFC (repeated-measures one-way ANOVA F3,24 = 31.80, P < 0.0001). (D) Representative example in one animal showing the evolution of PS amplitudes in the stimulation train at frequencies ranging from 5 to 40 Hz. Data represents the mean (solid line) and SEM (shaded area) of 10 repetitions for each frequency. (E) Quantification of the output spiking activity at all tested frequencies as the PS amplitude averaged across all delivered pulses, and normalized against 5 Hz. (Inset) Representative PS evoked by the first (black) and last (colored) pulse in the train for each stimulation frequency. Note the PS facilitation at 10 and 20 Hz (repeated-measures one-way ANOVA F3,12 = 8.06, P = 0.0007). (F) Linear correlation between BOLD signals in PFC and spiking activity in CA1 (r = 0.38, P < 0.0001). Multiple comparisons in all panels with Bonferroni’s test: *P < 0.05, **P < 0.01, ***P < 0.001, ****P < 0.0001. Asterisks color denotes comparison against 10 (yellow), 20 (green), or 40 (blue) Hz. Scale bars: 5 ms, 2 mV (C).

Figure 5. Frequency is the key parameter determining extrahippocampal activity propagation. (A) BOLD signal responses in the hippocampus and (B) PFC to different stimulation protocols varying the frequencies, number of pulses (p) and structure (bursts). Burst protocols consisted of 4 or 8 pulses separated by 250 or 600 ms, respectively. Both burst protocols produced the same result and has been pooled together for simplicity. Data points represent mean ± SEM (n = 3). Note the absence of extrahippocampal propagation at 40 Hz even if long (4 s) stimulation trains equivalent to those used at 10 Hz with 4 times the number of pulses are used. Note also the absence of propagation at higher frequencies (160 Hz) or when 40 Hz activation is patterned into theta- or delta-burst protocols. Repeated-measures One-way ANOVA for hippocampal (F4,8 = 1.9, P = 0.2) and PFC activations (F4,8 = 127.9, P < 0.0001). Multiple comparisons in all cases with Bonferroni’s test: ****P < 0.0001; NS (nonsignificant differences, P > 0.05).
LTP induction in the DG (Canals, Beyerlein et al. 2009) could be the result of 2 processes coordinated in the frequency domain (theta–beta range), (1) the enhanced transfer of activity from DG to CA3 (Yeckel and Berger 1998) and (2) the specific channeling to neocortical and striatal regions supported by the CA3 network as shown here. In the context of memory formation, it is tempting to hypothesize that changes of synaptic strength at particular network inputs (i.e., EC→DG) and the pacing of information coding in frequency bands favoring long-range hippocampal interactions (i.e., 10–20 Hz) may represent one possible routing instruction for hippocampal–neocortical interactions.

The present fMRI study draws attention, by virtue of observation of widespread neuronal activations, to important network implications of short-term synaptic plasticity. Whether long-term modulations of the same synapses have further functional consequences on the described frequency-dependent gating of activity propagation is an interesting question that requires further investigation.

**Systems Level**

Work over the past years points to cortical oscillations as a framework for organizing interactions among functionally specialized neurons in distributed brain networks (Singer 1993; Buzsaki 2006; Colgin and Moser 2010; Engel and Fries 2010; Lisman and Jensen 2013). For example, beta oscillations have recently been identified as a potential mechanism for synchronizing the evolution of neural representations in dispersed neural circuits after hippocampal-dependent learning (Igarashi et al. 2014). Similarly, theta rhythms have been proposed to coordinate hippocampal–prefrontal interactions in a spatial memory task (Jones and Wilson 2005; Siapas et al. 2005). Here, using a combination of fMRI and electrophysiological techniques, we demonstrate activity propagation to a distributed network of structures connected with the hippocampus at 10–20 Hz. Furthermore, long-range propagation at these frequencies to midline neocortical, prefrontal and sensory-motor networks together with ventromedial striatum (i.e., see Fig. 3B) is consistent with widespread beta oscillations recorded in those same regions commonly found both in sensory, attentional, and learning processes (Fries et al. 2001; Ravel et al. 2003; Martin et al. 2007; Berke et al. 2008; Colgin et al. 2009; Engel and Fries 2010; Howe et al. 2011; Igarashi et al. 2014). We are aware of the fundamental differences between recorded LFP signals oscillating at different frequencies and reflecting the orchestrated organization of multiple synaptic inputs (not necessarily matching the firing frequency of individual cells), and the activation frequencies experimentally imposed in our study by direct depolarizing pulses. We believe, however, based on the strong correspondence between the stimulation parameters used here and the temporal characteristics of action potential activity of hippocampal neurons observed in the behaving animal (Ranck 1973; Berger et al. 1983), that our results help to understand long-range interactions across neuronal populations. Although the specific range of relevant frequencies may vary for naturally occurring neuronal activations, the concept of frequency-dependent gating of activity propagation shown in our study and, most importantly, the unveiled polysynaptic routes of communication, would still be valid. Activity propagation segregated in the frequency domain most likely illustrates direct information transfer across brain regions, as discussed above, but may also represent a mechanism by which the CA3 output in the dorsal hippocampus coordinates the oscillatory framework that synchronizes activity across larger and functionally related territories. While a high level of coherence is a likely signature of long-range communication (von Stein and Sarnthein 2000), how such coherence is achieved mechanistically remains unclear. Overall, our results suggest that direct activity transfer at theta–beta frequencies between the hippocampus and connected structures may drive transient windows of strong functional coupling, organizing oscillations in dispersed networks and enhancing coherence as a consequence.

**Concluding Remarks**

Our results highlight the important role of short-term plasticity at CA3 synapses to route activity propagation and determine hippocampal–neocortical interactions. We show that activity paced at 10–20 Hz facilitates CA1 pyramidal cell output and promotes long-range interactions with a set of neocortical and striatal regions previously shown to be critically involved in memory formation.

**Supplementary Material**

Supplementary material can be found at: http://www.cercor.oxfordjournals.org

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**References**


