Gamma oscillations dynamically couple hippocampal CA3 and CA1 regions during memory task performance

Sean M. Montgomery and György Buzsáki*

Center for Molecular and Behavioral Neuroscience, Rutgers University, 197 University Avenue, Newark, NJ 07102

Edited by Fred H. Gage, The Salk Institute for Biological Sciences, San Diego, CA, and approved July 20, 2007 (received for review February 27, 2007)

The hippocampal formation is believed to be critical for the encoding, consolidation, and retrieval of episodic memories. Yet, how these processes are supported by the anatomically diverse hippocampal networks is still unknown. To examine this issue, we tested rats in a hippocampus-dependent delayed spatial alternation task on a modified T maze while simultaneously recording local field potentials from dendritic and somatic layers of the dentate gyrus, CA3, and CA1 regions by using high-density, 96-site silicon probes. Both the power and coherence of gamma oscillations exhibited layer-specific changes during task performance. Peak increases in the gamma power and coherence were found in the CA3–CA1 interface on the maze segment approaching the T junction, independent of motor aspects of task performance. These results show that hippocampal networks can be dynamically coupled by gamma oscillations according to specific behavioral demands. Based on these findings, we propose that gamma oscillations may serve as a physiological mechanism by which CA3 output can coordinate CA1 activity to support retrieval of hippocampus-dependent memories.

hippocampus | local field potential | retrieval | synchrony

Based primarily on lesion studies, a consensus has emerged that the hippocampal formation is critical for episodic memories (1–3). In accord with lesion studies, the firing patterns of hippocampal and entorhinal neurons exhibit prospective and retrospective coding of episodic information (4–7). However, the specific roles of the various hippocampal subregions (8) responsible for encoding, consolidation, and retrieval of memory traces have been long debated. Computational models (9–12) have postulated the autoassociative recurrent network of the CA3 region as a suitable substrate for storing memories, which could be subsequently recalled via replay from CA3 to CA1. Circumscribed lesions in animals (13, 14) and genetic manipulations (15) also support the view that the integrity of the CA3 region is crucial in memory retrieval, but little is known about the physiological mechanisms of CA3–CA1 coordination that might support this process.

Although recordings from multiple single neurons can assess the output representations of a network, the currently available large-scale unit recording methods do not have the ability to effectively monitor how information transfer is coordinated across several neuronal networks (16). On the other hand, although local field potentials (LFPs) lack single-neuron resolution, they reflect the temporal synchrony of local afferent activity and can effectively detect the changing modes of operation in local circuits (17–19). For example, synchronization of neuronal activity into coherent gamma-frequency oscillations can serve to bind representations (20, 21) and couple hippocampal and rhinal cortices during successful formation of declarative memories (22). To examine how regional networks within the hippocampal formation are dynamically coordinated during the retrieval process, we recorded LFPs from rats with high-density silicon probe arrays (23) during performance of a hippocampus-dependent delayed spatial alternation task on a modified T maze

(24). Because gamma oscillations in the hippocampus show clear regional and laminar patterns and the CA3 region is known to generate self-organized gamma oscillatory patterns (25–28), we examined the laminar-specific power and coherence of gamma oscillations. Our results show that gamma-frequency synchronization at the CA3–CA1 interface selectively increases on the center arm maze segment immediately preceding the T junction, independent of the motor aspects of task performance. These findings signify that gamma oscillations can dynamically coordinate hippocampal networks according to behavioral demands. Based on these results, we hypothesize that gamma oscillations may provide a physiological mechanism by which CA3–CA1 activity is coordinated to support retrieval of hippocampus-dependent memories.

Results

LFPs were recorded simultaneously in the dentate, CA3, and CA1 regions of the dorsal hippocampus by using a two-dimensional silicon probe array with 96 monitoring sites. In agreement with previous observations, the *in situ* recording sites in the various regions and layers could be determined with high spatial resolution ($\pm 30 \mu\text{m}$) by using a combination of spontaneous LFP patterns, multiple unit activity, evoked potentials in response to perforant path and commissural stimulation and posthoc histological identification of the anatomical position of each recording shank [Fig. 1; see supporting information (SI) Fig. 6A for electrode positions in all animals] (23, 29).

To engage hippocampal networks, rats ($n = 4$) were trained on a hippocampus-dependent, delayed spatial alternation task (Fig. 2A) (24). This task requires rats to encode their spatial response on each trial and, after a 10-second delay, retrieve this information to appropriately choose the opposite arm from the one entered on the previous trial. Although retrieval of previous trial information could occur in this task at any time before crossing the T junction, rats typically ran smoothly through the center arm and the T junction in one swift trajectory, suggesting that retrieval processes likely occur before reaching the T junction. All rats performed the task at high levels of proficiency ($>85\%$ correct).

Layer-Specific Gamma Power Increase on the Center Arm of the T Maze. We investigated the involvement of different hippocampal networks during performance of the spatial alternation task by

Author contributions: S.M.M. and G.B. designed research; S.M.M. performed research; S.M.M. analyzed data; and S.M.M. and G.B. wrote the paper.

The authors declare no conflict of interest.

This article is a PNAS Direct Submission.

Abbreviations: LFP, local field potential; str., stratum; GLM, general linear model; CSD, current source density.

*To whom correspondence should be addressed. E-mail: buzsa@rutgers.edu.

This article contains supporting information online at www.pnas.org/cgi/content/full/0701826104/DC1.

© 2007 by The National Academy of Sciences of the USA

Fig. 1. On-line calibration of recording sites. **A** 96-site silicon probe with recording pads spaced regularly over a 1.5 mm × 1.5 mm area is shown. The probe sites have a vertical spacing of 100 μ m and a horizontal spacing of 300 μ m. The anatomical position of recording electrodes in the different layers in the dentate gyrus and CA1 and -3 regions of the hippocampus was determined by using 1,1'-dioctadecyl-3,3,3',3'-tetramethylindocarbocyanine (DiI) labeling of electrode tracks (**A**) matched with known electrophysiological and anatomical characteristics of the hippocampus for each recording session. (**B** and **C**) Event-triggered averages of the LFP (traces) and CSD (color) centered on the estimated position from which the activity was recorded. (**B**) Ripple-triggered average responses (centered on ripple event, $n = 179$). Note the large sink (blue) associated with a sharp wave in CA1 str. radiatum. (**C**) Average evoked activity in response to perforant path stimulation (left aligned to stimulation, $n = 3$). Note the large sinks in the molecular layer of the dentate gyrus.

measuring the power (amplitude) of gamma oscillations simultaneously in various layers, across different portions of the task. In agreement with previous findings, maximum LFP gamma power was recorded in the hilar region of the dentate gyrus (25, 30). However, the relative power changes in the separate maze regions varied differentially across layers. As illustrated in Fig. 2 *B* and *C*, the amplitude of the gamma oscillation (40–120 Hz; *SI Fig. 7*), recorded from the middle of CA1 stratum (str.) radiatum, showed a selective increase on the center arm that was

not reliably observed in other hippocampal layers. Relative changes in gamma power were also observed in areas outside the CA1 str. radiatum in other segments of the maze, suggesting that the various hippocampal regions and afferents are involved in various task components (see *SI Movie 1*), but the present behavioral paradigm did not reliably isolate these events.

Because the running pattern of the rat was not identical in the different segments of the maze, and because previous research has shown speed and acceleration-dependent changes of hippocampal unit and field activity (31, 32), we first addressed the issue of whether overt motor behavior could account for the fluctuations in gamma power. Gamma power measurements were fit separately for each recording site with a general linear model (GLM), including as explanatory variables, running speed, acceleration, and maze region (see *Methods*). Similar to an analysis of covariance, GLM analysis dissects the trial-by-trial variance in gamma-power measurements that can be accounted for by the variables of interest (33, 34). GLM analysis showed that gamma power only weakly correlated with running speed and acceleration, as indicated by the consistently low r^2 values (“explained variance,” <10%) across all recording sites (Fig. 3*A*). Maze region, on the other hand, could explain >30% of the observed variance in gamma power at a number of sites. Especially striking was the degree to which the effect of maze region respected the laminar anatomy of the hippocampus, with the highest r^2 values confined to the CA1 str. radiatum. β values of the GLM analysis, showing the direction and magnitude of the change in gamma power for each maze region (Fig. 3*B*), revealed increased gamma power in CA1 str. radiatum on the center arm of the maze. Because the dominant projection to CA1 str. radiatum derives from the CA3 region (8), the increase of gamma power in this layer suggests an enhancement of CA3 output to CA1 on the center arm of the T maze.

To statistically compare task-related changes in the anatomical distribution of gamma power across animals, recording sites were assigned to specific layers (as shown in *SI Fig. 6B*), and the distribution of β values from each layer across all four rats was tested for reliable behavior-related changes (two-tailed t tests). Fig. 3*C* shows group statistics revealing differential increases in gamma power across hippocampal layers on the center arm versus other arms of the T maze. Although significant increases in gamma power were observed in several hippocampal layers on the center arm of the maze, the gamma power increase in CA1 str. radiatum was larger than in all other layers except for the CA3 pyramidal layer (ANOVA, Tukey post hoc test, $P < 0.05$). Further statistical analysis, including the effect of running speed, acceleration and all maze regions is shown in *SI Fig. 8*. We also tested whether gamma patterns were different on “error” versus “correct” trials (data not shown) but found no reliable difference, possibly because of the low number of error trials, high behavioral variability on error trials, and/or the foiled engagement of cognitive processes, e.g., retrieval mechanisms that return the wrong information.

Even though the center-arm increase in LFP gamma power shown in Fig. 3*C* was concentrated in specific layers, the LFP may be contaminated by volume conduction of activity in other layers. To reduce the contribution of possible volume-conducted fields, we first performed a one-dimensional current source density (CSD) analysis on the LFP data and repeated the GLM calculations on the derived data (Fig. 3*D*). This analysis revealed that the increased gamma oscillations in the CA3 pyramidal layer and CA1 str. radiatum were generated by local currents rather than volume conducted from the dentate gyrus or direct entorhinal input to the CA1 region (i.e., str. lacunosum-moleculare). CSD gamma power showed significant increases in the dentate molecular layer, possibly reflecting a change in entorhinal input, local circuitry, or CA3 back-projections to the inner molecular layer of the dentate gyrus (35). Gamma power also increased in

Task-Specific Enhancement of CA3–CA1 Gamma Synchrony. Although the GLM statistics are specifically designed to dissect individual explanatory variables (33), we performed additional control experiments to isolate the hypothesized mnemonic contribution to the observed changes in gamma oscillations. In addition to the alternation task, each rat was trained and tested on one of three control tasks (SI Fig. 10) possessing no memory requirement and no dependence on the hippocampus to control for motor and nonmnemonic cognitive aspects of task performance. Similar to the alternation task, in each control task the rat was held in a small delay area and then released to run for water reward. The initial segment of these control tasks showed similar speed and acceleration profiles to the center arm of the alternation task

Gamma oscillations have been suggested to assist various operations, including the binding of sensory attributes (20), synaptic plasticity (25), working memory (37, 38), sensory-motor coordination (21), attention (39), formation of cell assemblies (40), memory coding (22, 41, 42), and even consciousness (43, 44). These diverse putative functions indicate that gamma oscillations *per se* cannot be explicitly assigned to specific behavioral events. Instead, changes in the gamma oscillations recorded from a particular brain structure may be taken as an indicator that the supporting circuitry is engaged in a particular mode of computation.

Fig. 3. Gamma power on the center arm is maximally expressed in the CA3–CA1 associational system. Fit of gamma power with a GLM was used to assess contribution of various overt and covert variables. (A) r^2 of running speed and acceleration (Accel) and maze region. Note that the maze region explains substantial variance for CA1 str. radiatum recording sites, whereas speed and acceleration explains very little. (B) β values showing changes in LFP gamma power (in decibels) across the different maze regions for a single recording session. (C and D) Group statistics of gamma power changes on the center arm versus other arms across different anatomical layers (decibels, $\pm$ standard error; $P < 0.001$, Bonferroni corrected t tests) for LFP (C) and CSD (D). Also see SI Fig. 8.

CA3 and CA1 Networks Are Dynamically Coordinated by Gamma Oscillations. Gamma oscillations have diverse expression throughout the hippocampal networks. They are generated by a consortium of mechanisms and depend primarily on the activity of fast spiking basket neurons and GABA_A receptor-mediated inhibition and α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor-mediated excitation (19, 45–50). Gamma oscillations in the CA1 str. lacunosum-moleculare and the dentate gyrus are mainly under the control of entorhinal input (25, 51). However, evidence from *in vitro* and *in vivo* studies suggests that the CA3 region can generate gamma oscillations from the interactions between CA3 pyramidal cells and basket interneurons (25–27). The gamma phased-locked firing of CA3 projection neurons can in turn entrain the CA1 network (26, 36). We found dynamic changes in the power and coherence of CA3 and CA1 gamma oscillations. Some analyses also implicated specific layers in the dentate gyrus, although future experiments with dual entorhinal-hippocampal recordings will be needed to precisely determine to the extent to which these effects were related to direct entorhinal involvement, local activity, or CA3–dentate projections. Because our recordings principally sampled the CA3b and -c subregions, we cannot speculate about the role of the CA3a subregion from these data, given the differential wiring patterns of these subregions (35). Nevertheless, our results provide further support for a role of the CA3 region in generating self-organized gamma activity, independent of the

Fig. 4. CA3–CA1 gamma coherence is enhanced on the center arm. (A) Spatial distribution of gamma coherence between the CA3 pyramidal layer and example sites in CA1 str. radiatum (Upper; shown in B) and the dentate molecular layer (Lower). (B) Center-arm change in gamma coherence between a CA3 pyramidal reference site (circle) and all other hippocampal recording sites averaged over an entire recording session (20 trials). (C and D) Group data. (C) Change in gamma coherence between all hippocampal layer pairs on the center arm of the maze. Note that the peak increases in coherence are at the CA3–CA1 interface. (D) Bonferroni-corrected P values. Note that the most significant values are between the CA3 and CA1 layers. Also see SI Fig. 8.

gamma oscillations in other hippocampal regions. Additionally, our results provide evidence that the temporal coordination of CA3 and CA1 networks is dynamically regulated via gamma synchronization, according to behavioral demands.

Mnemonic Function of CA3–CA1 Gamma Coupling. Numerous lesion studies in humans and other animals link hippocampal networks to memory (2, 3). Previous computational models (9–11, 52), lesion studies (13, 14), and genetic knockout studies (15) have implicated CA3–CA1 communication in retrieval of hippocampus-dependent memories. We found a consistent pattern of changes in the power and coherence of gamma oscillations, showing increased synchrony in the CA3–CA1 interface on the center arm of the T maze. Using statistics and analysis of control tasks to rule out the contribution of overt behavior and other nonmnemonic variables, we found that the increased CA3–CA1 gamma coordination was specifically enhanced on the center arm of the T maze by cognitive aspects of alternation task performance. Performance of the delayed spatial alternation task requires, on each trial, that rats retrieve information about the previous journey to choose the opposite arm upon reaching the T junction. Although the exact spatial position(s) at which retrieval processes occur likely varies from trial to trial (as does the CA3–CA1 gamma synchrony increase seen in SI Movies 1 and 2), the fact that rats typically run through the center arm and the T junction in one swift trajectory suggests that retrieval and

1. Squire LR (1992) *Psychol Rev* 99:195–231.
2. Milner B, Squire LR, Kandel ER (1998) *Neuron* 20:445–468.
3. Eichenbaum H, Cohen NJ (2001) *From Conditioning to Conscious Recollection: Memory Systems of the Brain* (Oxford Univ Press, Oxford).
4. Wood ER, Dudchenko PA, Robitsek RJ, Eichenbaum H (2000) *Neuron* 27:623–633.
5. Frank LM, Brown EN, Wilson M (2000) *Neuron* 27:169–178.
6. Ferbinteanu J, Shapiro ML (2003) *Neuron* 40:1227–1239.
7. Shapiro ML, Ferbinteanu J (2006) *Proc Natl Acad Sci USA* 103:4287–4292.
8. Amaral DG, Witter MP (1995) in *The Rat Nervous System*, ed Paxinos G (Academic, New York), pp 443–490.
9. Marr D (1971) *Philos Trans R Soc London B* 262:23–81.
10. Treves A (1995) *J Comput Neurosci* 2:259–272.
11. Rolls ET (1996) *Hippocampus* 6:601–620.
12. Wallenstein GV, Eichenbaum H, Hasselmo ME (1998) *Trends Neurosci* 21:317–323.
13. Brun VH, Otnass MK, Molden S, Steffenach HA, Witter MP, Moser MB, Moser EI (2002) *Science* 296:2243–2246.
14. Steffenach HA, Sloviter RS, Moser EI, Moser MB (2002) *Proc Natl Acad Sci USA* 99:3194–3198.
15. Nakazawa K, Quirk MC, Chitwood RA, Watanabe M, Yeckel MF, Sun LD, Kato A, Carr CA, Johnston D, Wilson MA, et al. (2002) *Science* 297:211–218.
16. Buzsáki G, Draguhn A (2004) *Science* 304:1926–1929.
17. Bressler SL, Kelso JA (2001) *Trends Cognit Sci* 5:26–36.
18. Varela F, Lachaux JP, Rodríguez E, Martinerie J (2001) *Nat Rev Neurosci* 2:229–239.
19. Bartos M, Vida I, Jonas P (2007) *Nat Rev Neurosci* 8:45–56.
20. Gray CM, König P, Engel AK, Singer W (1989) *Nature* 338:334–337.
21. Engel AK, Fries P, Singer W (2001) *Nat Rev Neurosci* 2:704–716.
22. Fell J, Klaver P, Lehnertz K, Grunwald T, Schaller C, Elger CE, Fernandez G (2001) *Nat Neurosci* 4:1259–1264.
23. Csicsvari J, Henze DA, Jamieson B, Harris KD, Sirota A, Bartho P, Wise KD, Buzsáki G (2003) *J Neurophysiol* 90:1314–1323.
24. Ainge JA, van der Meer MA, Langston RF, Wood ER (June 6, 2007) *Hippocampus*, 10.1002/hipo.20301.
25. Bragin A, Jando G, Nadasdy Z, Hetke J, Wise K, Buzsáki G (1995) *J Neurosci* 15:47–60.
26. Csicsvari J, Jamieson B, Wise KD, Buzsáki G (2003) *Neuron* 37:311–322.
27. Mann EO, Paulsen O (2005) *Prog Biophys Mol Biol* 87:67–76.
28. Isomura Y, Sirota A, Ozen S, Montgomery S, Mizuseki K, Henze DA, Buzsáki G (2006) *Neuron* 52:871–882.
29. Bragin A, Jando G, Nadasdy Z, van Landeghem M, Buzsáki G (1995) *J Neurophysiol* 73:1691–1705.
30. Stumpf C (1965) *Electroencephalogr Clin Neurophysiol* 18:477–486.
31. Whishaw IQ, Vanderwolf CH (1973) *Behav Biol* 8:461–484.
32. McNaughton BL, Barnes CA, O'Keefe J (1983) *Exp Brain Res* 52:41–49.
33. Rencher AC (2002) *Methods of Multivariate Analysis* (Wiley, New York).
34. Jacobs J, Hwang G, Curran T, Kahana MJ (2006) *NeuroImage* 32:978–987.
35. Li XG, Somogyi P, Ylinen A, Buzsáki G (1994) *J Comp Neurol* 339:181–208.
36. Fisahn A, Pike FG, Buhl EH, Paulsen O (1998) *Nature* 394:186–189.
37. Lisman JE, Idiart MA (1995) *Science* 267:1512–1515.
38. Howard MW, Rizzuto DS, Caplan JB, Madsen JR, Lisman J, Aschenbrenner-Scheibe R, Schulze-Bonhage A, Kahana MJ (2003) *Cereb Cortex* 13:1369–1374.
39. Fell J, Fernandez G, Klaver P, Elger CE, Fries P (2003) *Brain Res Brain Res Rev* 42:265–272.
40. Harris KD, Csicsvari J, Hirase H, Dragoi G, Buzsáki G (2003) *Nature* 424:552–556.
41. Sederberg PB, Kahana MJ, Howard MW, Donner EJ, Madsen JR (2003) *J Neurosci* 23:10809–10814.
42. Sederberg PB, Schulze-Bonhage A, Madsen JR, Bromfield EB, McCarthy DC, Brandt A, Tully MS, Kahana MJ (2007) *Cereb Cortex* 17:1190–1196.
43. Llinas R, Ribary U, Contreras D, Pedraarena C (1998) *Philos Trans R Soc London B* 353:1841–1849.
44. Sauve K (1999) *Conscious Cognit* 8:213–224.
45. Buzsáki G, Leung LW, Vanderwolf CH (1983) *Brain Res* 287:139–171.
46. Whittington MA, Traub RD, Jefferys JG (1995) *Nature* 373:612–615.
47. Whittington MA, Traub RD, Kopell N, Ermentrout B, Buhl EH (2000) *Int J Psychophysiol* 38:315–336.
48. Wang XJ, Buzsáki G (1996) *J Neurosci* 16:6402–6413.
49. Traub RD, Whittington MA, Colling SB, Buzsáki G, Jefferys JG (1996) *J Physiol (London)* 493:471–484.
50. Traub RD, Bibbig A, LeBeau FE, Buhl EH, Whittington MA (2004) *Annu Rev Neurosci* 27:247–278.
51. Chrapak S, Pare D, Llinas R (1995) *Eur J Neurosci* 7:1548–1557.
52. Hasselmo ME, Bodelon C, Wyble BP (2002) *Neural Comput* 14:793–817.
53. Bower MR, Euston DR, McNaughton BL (2005) *J Neurosci* 25:1313–1323.
54. Lisman JE (1997) *Trends Neurosci* 20:38–43.
55. Buzsáki G, Traub RD, Pedley TA (2003) in *Current Practice of Clinical Encephalography*, eds Ebersole JS, Pedley TA (Lippincott Williams & Wilkins, Philadelphia), pp 1–11.
56. Robbe D, Montgomery SM, Thome A, Rueda-Orozco PE, McNaughton BL, Buzsáki G (2006) *Nat Neurosci* 9:1526–1533.
57. Sayer RJ, Friedlander MJ, Redman SJ (1990) *J Neurosci* 10:826–836.