

6. X. Yang *et al.*, *EMBO J.* **18**, 1280 (1999).
7. Y. Zhu *et al.*, *Cell* **94**, 703 (1998).
8. R. Derynck, R. J. Akhurst, A. Balmain, *Nature Genet.* **29**, 117 (2001).
9. C. Dong *et al.*, *Mol. Cell* **5**, 27 (2000).
10. V. Bennett, A. J. Baines, *Physiol. Rev.* **81**, 1353 (2001).
11. W. J. Nelson, P. J. Veshnock, *J. Cell Biol.* **103**, 1751 (1986).
12. M. A. De Matteis, J. S. Morrow, *J. Cell Sci.* **113**, 2331 (2000).
13. L. Mishra *et al.*, *Oncogene* **18**, 353 (1999).
14. M. Weinstein *et al.*, *Mol. Cell Biol.* **21**, 5122 (2001).
15. L. Mishra *et al.*, *Int. J. Dev. Biol.* **42**, 221 (1998).
16. Materials and methods are available as supporting material on Science Online.
17. J. Larsson *et al.*, *EMBO J.* **20**, 1663 (2001).
18. E. Piek *et al.*, *J. Cell Sci.* **112**, 4557 (1999).
19. E. Piek *et al.*, *J. Biol. Chem.* **276**, 19945 (2001).
20. Y. Tang *et al.*, data not shown.
21. C. D. Bhanumathy *et al.*, *Dev. Dyn.* **223**, 59 (2002).
22. A. Sasaki *et al.*, *J. Biol. Chem.* **276**, 17871 (2001).
23. A. Moustakas, C.-H. Heldin, *Genes Dev.* **16**, 1867 (2002).
24. L. Jayaram, J. Massague, *J. Biol. Chem.* **275**, 40710 (2000).
25. We thank C. Li for invaluable assistance with generating mutant *elf* mice and mouse blastocyte injection; S. J. Kim, A. Roberts, J. Wrana, L. Attisano, J. Massague, and A. Hata for the kind gift of Smad expression vectors; S. Danovitch, E. P. Reddy, and M. Zaslloff for their constructive criticism during the evolution of this manuscript; and R. Redman, E.

Fiores, A. Lorman, and E. Mishra for assistance with immunohistochemistry. Supported by NIH R01 DK56111 (L.M.), NIH R01 DK58637 (B.M.), a Veterans Administration merit award (L.M.), NIH R03 DK53861 (L.M.), the Betty and Harry Myerberg Foundation, and the Elisabeth and John Cox Foundation. This manuscript is dedicated to J.B.

#### Supporting Online Material

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Materials and Methods  
Figs. S1 to S3  
References

10 July 2002; accepted 6 December 2002

## Dynamics of the Hippocampus During Encoding and Retrieval of Face-Name Pairs

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The medial temporal lobe (MTL) is critical in forming new memories, but how subregions within the MTL carry out encoding and retrieval processes in humans is unknown. Using new high-resolution functional magnetic resonance imaging (fMRI) acquisition and analysis methods, we identified mnemonic properties of different subregions within the hippocampal circuitry as human subjects learned to associate names with faces. The cornu ammonis (CA) fields 2 and 3 and the dentate gyrus were active relative to baseline only during encoding, and this activity decreased as associations were learned. Activity in the subiculum showed the same temporal decline, but primarily during retrieval. Our results demonstrate that subdivisions within the hippocampus make distinct contributions to new memory formation.

Structures within the MTL play a crucial role in forming new associations or episodic memories. Memory formation is a dynamic process: As new information becomes better learned, the hippocampus appears to be less critical (1, 2). The complex architecture of the hippocampus would seem to orchestrate this transition (3). Several studies have demonstrated some degree of subregion specificity within the hippocampus and related structures. In particular, recognition memory may require the perirhinal cortex (4), spatial memory may depend on the parahippocampal cortex (5, 6), and encoding and retrieval may involve the anterior and posterior hippocampus, respectively (7, 8). However, no studies to date have directly examined

how activity patterns within different substructures of the MTL change during learning. Here, we use functional magnetic resonance imaging (fMRI) in human volunteers to identify changes in the blood oxygen level–dependent (BOLD) response, reflecting neural activity, within different substructures of the MTL, as subjects progressively learn new associations.

Imaging the medial temporal subregions is technically challenging: Not only are the individual structures quite small, but the hippocampus itself is rolled into a compact spiral, making it difficult to isolate activity within any one region on the planar sections acquired in MRI scans. In order to parcel out neural activity in the subregions, we developed techniques to acquire high-resolution structural (0.4 by 0.4 mm) and functional (1.6 by 1.6 mm) MRI data and to localize functional activity precisely within the substructures of the hippocampus by “unfolding” the hippocampal cortex, revealing the entirety of each hippocampal subregion [CA fields 1, 2, and 3; dentate gyrus (DG); and subiculum] and adjacent neocortical regions (parahippocampal, entorhinal, perirhinal, and fusiform) in a single plane, or “flat map” (9–11) (Fig. 1). Briefly, this procedure begins by

first demarcating the boundaries between the architectonic subregions on the high-resolution structural MR images (Fig. 1A) and then segmenting and separating out the white matter and CSF throughout the MTL, retaining only the gray matter sheath (Fig. 1B). We then computationally extract and flatten the gray matter volume (similar to flattening the globe into a flat map of the world) and project the demarcated boundaries to produce unfolded flattened maps of the hippocampus (Fig. 1C). Because subjects vary in the anatomy of their MTLs, we constructed a template representing the typical anatomy of our subject population by averaging together the individual demarcation boundaries across subjects (12). Computational warping techniques transform an individual subject’s hippocampal maps to the flat hippocampal template space (13). The same transformation parameters are then applied to the coregistered functional MRI scans, which delivers high-resolution fMRI data in a standardized flat space. This procedure enabled us to measure activity over time in each subregion (e.g., the combined CA 2, 3, and DG termed “CA2,3DG,” CA 1, subiculum, fusiform, etc.) and to perform powerful group statistics across subjects.

Using this method, we scanned ten subjects while they performed a face-name association task in which a series of unfamiliar faces were paired with names (11). Learning the names of new faces is an essential aspect of everyday human memory that is known to engage the hippocampus (14, 15). We separated encoding blocks, where subjects saw the faces with the names and tried to commit them to memory, from recall blocks, where subjects saw the faces only and had to generate the correct name (Fig. 2). A distraction task prevented rote rehearsal between encoding and recall blocks. Subjects viewed the same face-name combinations four times so that the information was well learned by the last trial.

The subjects exhibited a positive learning curve over time as displayed in Fig. 3B. To determine the amount of new information successfully encoded on a given trial, we

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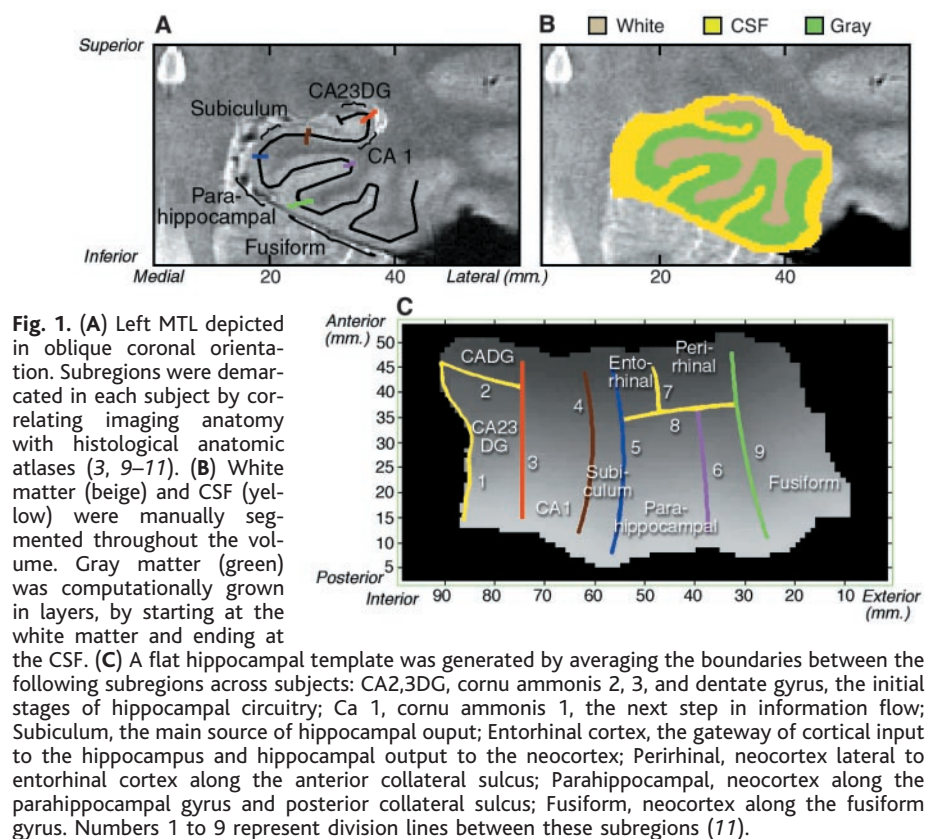
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calculated in each block the number of associations that subjects had encoded and recalled successfully for the first time (11) (Fig. 3B). This curve represents how many new successes at learning and retrieving occurred in each learn and recall block, and it reflects the total amount of novel and successful mnemonic processing that has been found to correlate with MTL activity in the literature (9, 10, 16–19). Using this group-averaged incremental performance curve, we regressed MR signal intensity in each pixel and each subject with two waveforms reflecting either performance during learning or performance during retrieval, and we then statistically tested whether the slope of each regression for a given pixel was on-average different from zero (Fig. 3A) (11, 20). Strikingly, changes in the fMRI signal from baseline in the anterior CA 2 and 3 fields and DG closely followed this learning curve, but only during the encoding trials (Fig. 3A, Learn) and not during the interspersed retrieval blocks (Fig. 3A, Recall). We verified this by blindly selecting all voxels in the bilateral CA2,3DG regions in each subject and measuring the time course of the fMRI signal to see how closely it conformed to that of our a priori hypothesis (11). Random effects analysis of the subjects' time courses confirmed that CA2,3DG fMRI signal change correlated with the group-averaged incremental learning curve during encoding ( $P = 0.012$ ) but not during retrieval ( $P = 0.478$ ), and the difference between learning and retrieval was significant ( $P = 0.006$ ) (Fig. 3B). The lack of fMRI signal increase in the recall blocks suggests that there is little or no change in activation relative to the control task during retrieval. The large increases in signal in the encode blocks implies a relatively large increase in cerebral blood flow, which correlates with neural activity, during encoding (21). Thus, fields CA2,3DG appeared to be more active during encoding than during retrieval of new face-name associations.

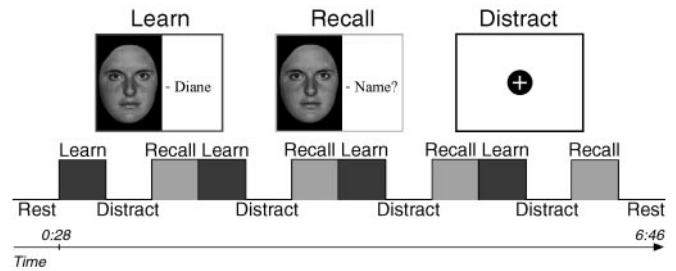
In contrast, fMRI signal change in the posterior subiculum correlated with the incremental learning curve during retrieval ( $P = 0.022$ ) but not during encoding ( $P = 0.250$ ). This difference between learning and retrieval ( $P = 0.341$ ) was not significant, however, probably because of the presence of sub-threshold subicular activity during encoding (Fig. 3B). The subiculum thus seems to be most active compared with baseline during retrieval, and relatively less active during encoding. A three-way ANOVA of region (CA2,3DG versus subiculum) by condition (encoding versus retrieval) by subject revealed a significant interaction between region and condition ( $P = 0.010$ ), confirming the double dissociation of activation patterns in CA2,3DG and the subiculum (22).

Prior studies have found increases in hip-



**Fig. 1.** (A) Left MTL depicted in oblique coronal orientation. Subregions were demarcated in each subject by correlating imaging anatomy with histological anatomic atlases (3, 9–11). (B) White matter (beige) and CSF (yellow) were manually segmented throughout the volume. Gray matter (green) was computationally grown in layers, by starting at the white matter and ending at the CSF. (C) A flat hippocampal template was generated by averaging the boundaries between the following subregions across subjects: CA2,3DG, cornu ammonis 2, 3, and dentate gyrus, the initial stages of hippocampal circuitry; Ca 1, cornu ammonis 1, the next step in information flow; Subiculum, the main source of hippocampal output; Entorhinal cortex, the gateway of cortical input to the hippocampus and hippocampal output to the neocortex; Perirhinal, neocortex lateral to entorhinal cortex along the anterior collateral sulcus; Parahippocampal, neocortex along the parahippocampal gyrus and posterior collateral sulcus; Fusiform, neocortex along the fusiform gyrus. Numbers 1 to 9 represent division lines between these subregions (11).

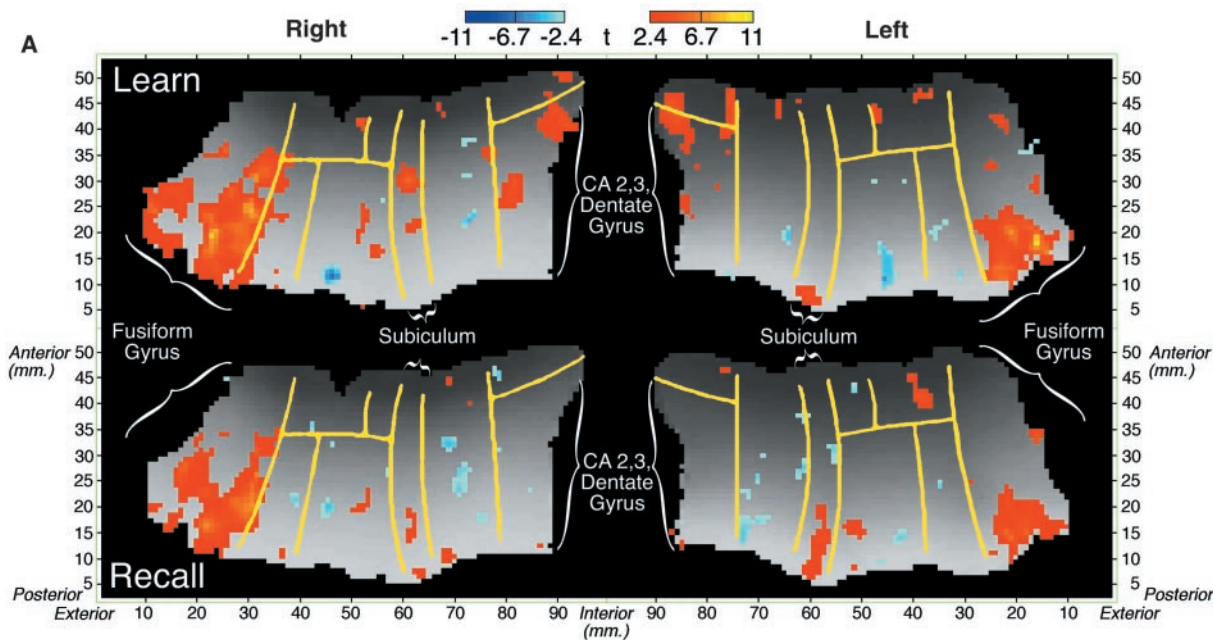
**Fig. 2.** Subjects serially studied eight pairs of faces and names during four learning blocks and attempted to recall the name when presented with the same eight faces during four interleaved retrieval blocks (11). A distraction task required subjects to press a button when a fixation cross changed to a circle.



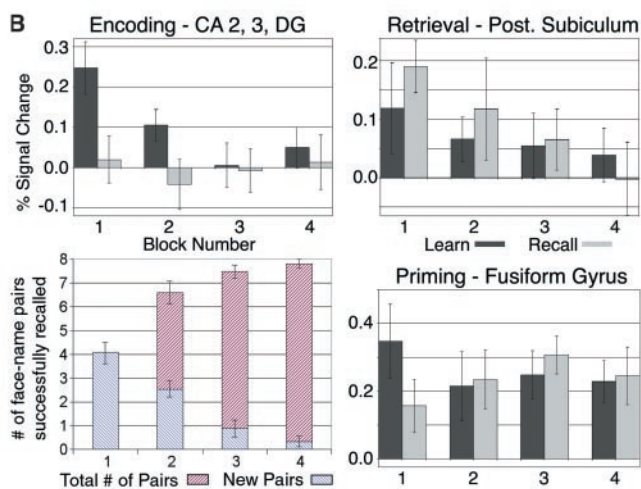
poampal activity when subjects process novel stimuli (16). The decline in activity in CA2,3DG during encoding could be explained by decreasing stimulus novelty. However, if activity in the CA 2 and 3 and DG simply reflected novelty, a similar pattern of activity would be expected during retrieval, because of the presentation of novel faces not fully encoded. Because not even a trend of such activity was observed ( $P = 0.478$ ), stimulus novelty cannot account for our results in the CA2,3DG. Similarly, novelty processing cannot explain the difference in activity patterns during encoding and retrieval that we observed in the subiculum. The fusiform gyrus, which is implicated in perceptual face processing (23), does exhibit activity consistent with novelty effects. Here, we found an initial decrement in fusiform activity after the first encoding block, consis-

tent with perceptual priming, the phenomenon in which repetition of a visual stimulus results in decreased activity (24). Thereafter, activity remained constant throughout both learning and recall (Fig. 3B). Thus, although the fusiform gyrus was active during the memory task ( $P = 0.0001$ ), the activation pattern over time was unrelated to the learning of associations and instead heightened for novel faces. To illustrate the spatial distribution of function across subregions in three dimensions, we superimposed activity during learning and recall on a reconstruction of the left MTL (Fig. 4) (11). Laterality effects were not observed in either learning or recall (11).

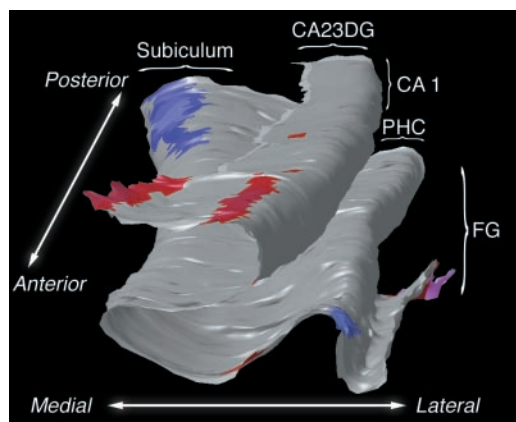
These results demonstrate a strong, parametric correlation between CA2,3DG activity and the storage of new associations; as the number of new associations learned decreased from block to block, activity in these regions



**Fig. 3.** (A) Random effects  $t$  maps ( $t \geq 2.4$ , statistical maps of significantly activated regions as seen by fMRI) for incremental performance during learning (top) and recall (bottom). (B) The averaged signal changes during each of the task blocks for all voxels in the bilateral CA2,3DG, posterior subiculum, and fusiform gyri (see Fig. 2), without any pixel selection (11). Percent signal change is relative to the adjacent baseline blocks (rest and distraction conditions), and error bars correspond to the standard error across subjects ( $n = 10$ ). At the bottom left, absolute and incremental recall performance are displayed across blocks.



**Fig. 4.** Encoding pixels shown in red, recall pixels in blue, and overlapping in purple, shown on a rendered left MTL representative of the average anatomy of our subject pool (11).



fell in parallel. We found a similarly strong relationship between activity in the subiculum and retrieval of newly learned associations. Our results present challenges for existing computational models of MTL function (25,

26). It is possible that incorporating aspects of the anatomy that are specific to either the components of CA23DG or especially the subiculum could explain the dissociation of functions that we observe here.

It is noteworthy that none of the MTL regions showed activity that reliably correlated with the total number of associations retrieved, which contains both newly learned items and previously recalled pairs. Our first experiment could not examine areas outside of the MTL, however, because our scan planes were restricted in order to maximize spatial resolution. To identify areas in the cortex correlated with the retrieval of both new and practiced associations, we performed whole-brain scanning on seven of the same subjects during the same paradigm with unique stimuli (0). Analysis revealed that the left anterior prefrontal cortex (PFC) exhibited activity that increased over time in proportion to the total number of names the subject had learned (fig. S1). The PFC is strongly implicated in memory retrieval and, particularly, in retrieval effort or retrieval success (27–30). Increased activity in PFC may reflect the greater number of successful retrievals as

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subjects learn the face-name associations. The pattern in PFC contrasts with the decreases in activity observed in the hippocampus over repeated retrieval blocks. As retrieval becomes practiced, it may be performed more efficiently, and require less activity in the hippocampus (31). Several other brain regions, including left posterior superior temporal gyrus, right lateral posterior fusiform, left ventral occipital cortex, and left motor areas also tracked retrieval success (11), which suggests that a network of linguistic and perceptual areas cooperate with anterior PFC in memory retrieval.

In any given cognitive task, encoding and retrieval may be difficult to separate completely. For example, retrieval operations likely occurred during our later encoding blocks, as subjects are presented face-name pairs they have already learned. Despite the presence of mixed processes in each task, our ability to reliably dissociate encoding and retrieval across regions most likely reflects the predominance of one process. Attention is another cognitive process whose effects could have influenced our results. The decreases in activity that we observed could be because subjects paid less attention to the tasks as they became easier. Changes in attention, however, cannot explain the dissociation that we observed between regions in the MTL or why the decline in activity in subiculum during retrieval is accompanied by an increase in activity in PFC.

Most prior imaging studies of declarative memory have examined a relatively static "snap-shot" of the memory system obtained after a single learning episode per item (16–19, 28–30). By measuring changes in cortical activity as declarative learning progressed, we found evidence for two dissociations within the

neural substrates of memory: Encoding and retrieval appear to engage different regions within the hippocampus preferentially, and practice appears to reduce demands on the hippocampus while increasing activity in neocortex related to retrieval success. Our examination of hippocampal activity during the learning process has revealed some of the cortical dynamics that support the acquisition of new information.

### References and Notes

1. L. Squire, *Psychol. Rev.* **99**, 195 (1992).
2. R. Clark, N. Broadbent, S. Zola, L. Squire, *J. Neurosci.* **22**, 4663 (2002).
3. D. Amaral, R. Insausti, in *The Human Nervous System*, G. Praxinos, Ed. (Academic Press, San Diego, 1990), pp. 711–755.
4. J. Aggleton, M. Brown, *Behav. Br. Sci.* **22**, 425 (1999).
5. V. Bohbot et al., *Neuropsychologia* **36**, 1217 (1998).
6. C. Ploner et al., *Cereb. Cortex* **10**, 1211 (2000).
7. M. Lepage, R. Habib, E. Tulving, *Hippocampus* **8**, 313 (1998).
8. D. Schacter, A. Wagner, *Hippocampus* **9**, 7 (1999).
9. M. Zeineh, S. Engel, S. Bookheimer, *NeuroImage* **11**, 668 (2000).
10. M. Zeineh, S. Engel, P. Thompson, S. Bookheimer, *Anat. Rec. (New Anat.)* **265**, 111 (2001).
11. Materials and methods are available as supporting material in *Science* Online.
12. P. Thompson, R. Woods, M. Mega, A. Toga, *Hum. Brain Mapp.* **9**, 81 (2000).
13. P. Thompson, A. Toga, in *Brain Warping*, A. W. Toga, Ed. (Academic Press, San Diego, 1998), pp. 311–336.
14. R. Sperling et al., *Hum. Brain Mapp.* **14**, 129 (2001).
15. J. Crane, B. Milner, *Neuropsychologia* **40**, 530 (2002).
16. C. Stern et al., *Proc. Natl. Acad. Sci. U.S.A.* **93**, 8660 (1996).
17. J. B. Brewer, Z. Zhao, J. E. Desmond, G. H. Glover, J. D. E. Gabrieli, *Science* **281**, 1185 (1998).
18. A. D. Wagner et al., *Science* **281**, 1188 (1998).
19. L. Eldridge, B. Knowlton, C. Furmanski, S. Bookheimer, S. Engel, *Nature Neurosci.* **3**, 1149 (2000).
20. An average group curve was used for all statistical analyses. The standard deviation in performance across subjects was 1.03, 0.981, 0.532, and 0.472 for recall blocks 1 to 4, respectively. Analyses conducted with individual curves produced learning and retrieval results identical to those presented here.
21. N. Logothetis, J. Pauls, M. Augath, T. Trinath, A. Oeltermann, *Nature* **412**, 150 (2001).
22. *P* values reflect random effects analyses on ROIs based on anatomical, not statistical, criteria, uncorrected for multiple comparisons. Our prior work has enabled us to hypothesize the importance of the hippocampus proper to the memory task in this experiment (9, 19). Nonetheless, our key result, the interaction between subregion (CA 2 and 3, and DG versus subiculum) and task (encoding versus retrieval) would survive Bonferroni correction of the three regions of the hippocampus proper ( $P = 0.03$ ).
23. N. Kanwisher, J. McDermott, M. Chun, *J. Neurosci.* **17**, 4302 (1995).
24. R. Buckner et al., *Neuron* **20**, 285 (1998).
25. E. Rolls, *Hippocampus* **6**, 601 (1996).
26. R. O'Reilly, J. Rudy, *Psychol. Rev.* **108**, 311 (2001).
27. D. Donaldson, S. Petersen, J. Ollinger, R. Buckner, *NeuroImage* **13**, 129 (2001).
28. M. Rugg, P. Fletcher, P. Chua, R. Dolan, *NeuroImage* **10**, 520 (1999).
29. R. Buckner et al., *NeuroImage* **7**, 163 (1998).
30. K. McDermott, T. Jones, S. Petersen, S. Lageman, H. Roediger, *J. Cogn. Neurosci.* **12**, 965 (2000).
31. The pattern of activity observed here suggests that the subiculum is initially critical for memory retrieval, but that additional learning allows this function to be performed with less engagement of hippocampal circuitry. These results may be consistent with models that propose a specific role for the hippocampus in supporting episodic memory retrieval (7, 19, 25, 26); as items are practiced they may lose their link to a specific memory context and become less episodic. The process observed here, however, differs from classical memory consolidation (32), which takes a much greater amount of time and so could not be observed directly in this experiment. Possible links between memory consolidation and loss of episodic content remain a topic of investigation.
32. J. L. McGaugh, *Science* **287**, 248 (2000).
33. This work was supported by NIMH NRSA MH12167, NIH MSTP GM08042, NIH AG 13308, the UCLA Affiliates, the Brain Mapping Medical Research Organization, the Ahmanson Foundation, the Pierson-Lovelace Foundation, the Tamkin Foundation, and the Jennifer Jones-Simon Foundation. We thank B. Wandell for use of his unfolding software and J. Mazziotta, R. Poldrack, and B. Knowlton for their comments on this manuscript.

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Materials and Methods  
Fig. S1

27 August 2002; accepted 12 December 2002

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