The Parahippocampus Subserves Topographical Learning in Man

The hippocampus has been proposed as the site of neural representation of large-scale environmental space, based upon the identification of place cells (neurons with receptive fields for current position in the environment) within the rat hippocampus and the demonstration that hippocampal lesions impair place learning in that area. The inability to identify place cells within the monkey hippocampus and the observation that unilateral hippocampal lesions do not selectively impair topographic behavior in humans suggest that alternate regions may subserve this function in man. To examine the contribution of the hippocampus and adjacent medial-temporal lobe structures to topographic learning in the human, a 'virtual' maze was used as a task environment during functional magnetic resonance imaging studies. During the learning and recall of topographic information, medial-temporal activity was confined to the parahippocampal gyrus. This activity accords well with the lesion site known to produce topographical disorientation in humans. Activity was also observed in cortical areas known to project to the parahippocampus and previously proposed to contribute to a network subserving spatially guided behavior.

The neural basis of allocentric (environment-centered) spatial learning and representation has been studied for several decades with lesion and single-unit recording studies in the rodent. These investigations have revealed that several different neuro-anatomical areas are involved in the transformation of spatial information from its initial retinotopic form into a dynamic representation of place. Most significantly, in 1971 O'Keefe and Dostrovsky reported the presence of a population of 'place cells' within the rat hippocampus that fire preferentially when the animal is in a specific location within its environment. This finding led to the proposal that the hippocampus maintains a 'cognitive map' of learned environments, with the pattern of activity indicating the current position within that space (O'Keefe and Nadel, 1978). Additional evidence regarding the importance of the hippocampus in topographic learning was provided by the report of Morris and colleagues (1982) that rats with hippocampal lesions were impaired on a specific test of place learning, the water maze task.

Attempts to demonstrate the importance of medial-temporal structures for topographical behavior in the primate have met with greater challenges, primarily because of the increased scale upon which these behaviors are conducted in the human and monkey. While numerous lesion (see e.g. George et al., 1989; Baylis and Moore, 1994) and neurophysiology (Cahusac et al., 1989; Miyashita et al., 1989) experiments have examined 'spatial' tasks in general, these studies have generally not captured the allocentric frame of reference crucial to topographic representation. However, recent studies by Rolls, O'Mara and their colleagues have examined hippocampal neuronal activity during whole body motion in the restrained (O'Mara et al., 1994; Rolls and O'Mara, 1995) and freely moving (Rolls et al., 1995) monkey. While these studies have identified neurons with clearly spatial functions, including those responsive to the location of the monkey's gaze and whole body motion, 'place cells' per se have not been identified. Additionally, clinical case reports have shown that unilateral lesions of the hippocampus proper in human patients do not impair topographic memory (deRenzi, 1982; Hublet and Demeurisse, 1992). These findings suggest that hippocampal function in spatial cognition may differ substantially between the rodent and primate.

The anatomically closely linked cortical areas that surround the hippocampus, including the entorhinal, parahippocampal and perirhinal gyri, have also been examined with regard to their memory function. Lesions of the rat parahippocampal gyrus (which encompasses the entorhinal and perirhinal cortex in the rat) also cause deficits in place learning (Schenk and Morris, 1985; Wilg and Bilkey, 1994; Nagahara et al., 1995), and similar lesions in the pigeon disrupt local homing behavior (Bingman and Mench, 1990).

These extra-hippocampal areas, in addition to the hippocampus proper, have been identified as important elements in human and non-human primates for declarative memory, the representation of information in a flexible form (Squire, 1992). In the monkey, parahippocampal and perirhinal lesions in conjunction with hippocampal lesions have been shown to produce a greater memory deficit on amnesia-sensitive tasks, such as delayed-match-to-sample and object retention, than hippocampal lesions alone (Zola-Morgan et al., 1993; Alvarez et al., 1995). Lesions of the perirhinal and parahippocampal areas also produce memory deficits equal to or greater than those produced by hippocampal lesions alone (Zola-Morgan et al., 1989; Suzuki et al., 1993).

In an effort to address the conflicting evidence regarding the regional function of the medial temporal lobes in the rodent and primate we have used functional magnetic resonance imaging (fMRI) to localize the neural substrates of human topographic spatial learning within the hippocampal system. A 'virtual-reality' environment was used in order to allow subjects to engage in allocentric learning of a spatially extended place. Differences in activity observed while subjects explored a complex maze, as compared with a control environment with scant topographic detail, were hypothesized to reveal the neural basis of the acquisition of representations of extended environments. Because both conditions expose the subject to a virtual place, functional activity was expected to identify primarily those areas contributing to topographic learning, as opposed to those areas which activate only in response to motion within, or perception of, real-world environments.

Materials and Methods
We studied nine right-handed males, aged 24-34 years. All subjects provided informed, written consent.
Experimental Design

The application of neuroimaging techniques, which require subject immobility, to investigate the cognition of spatial exploration, which demands subject motion, presents a unique experimental challenge. We have addressed this difficulty by creating a simple ‘virtual-reality’ environment which subjects are able to explore. This approach makes the assumption that the virtual-reality experience is sufficiently realistic to engage the same cognitive mechanisms, both behavioral and neural, that are employed in similar real-world tasks. Within the environment subjects performed tasks requiring them to acquire and utilize topographic information.

Virtual Environment

We modified a commercially available computer game to create a virtual-reality maze (Wolf 3D by ID Software and MacPlay, maps generated using WolfEdit 1.1, a freeware program by G. C. Ewing). This software presents a first-person point of view, three-dimensional, fully textured environment. Figure 1A depicts a typical view experienced by the subject within the maze. The subject’s view of the maze from within the magnet bore was in full color and subtended a 24° horizontal and 8° vertical visual angle. Stimuli were projected upon a screen which the subject viewed through a mirror. The subject controlled his movement through the environment with the use of a four-button, fiber-optic control pad operated with both thumbs.

The maze itself was of simple design, with three points of three-way intersection, five cul-de-sacs and multiple non-intersection turns (figure 1B). Each cul-de-sac contained a different object, one of which served to mark the subject’s point of entry into the maze. All intersections appeared identical when approached from different directions, preventing the subject from learning routes based solely upon regional landmarks.

Testing Protocol

Figure 2 depicts the overall experimental design. Functional data were acquired while subjects attempted to learn [explore] the topographical arrangement of the maze and again while subjects performed retrieval [navigation] tasks within the now familiar maze. Subjects were instructed to freely explore the maze during learning. Periodically, scanning was stopped and subjects attempted to draw a map, requiring them to translate their primary visual and motor experience into an aerial-view, topographic representation of their environment. The map was judged for accurate depiction of geometric detail and correct placement of cul-de-sac objects.

After demonstrating familiarity with the maze, subjects performed a retrieval task which required them to utilize their learned representation. During the retrieval task the subject entered the maze at the location of one of the four objects previously visited and travelled back and forth between this point and the maze start as quickly as possible. This navigation attempt was repeated for each of the four objects, preventing
the subject from memorizing a rote sequence of movements which would suffice to traverse the path.

All scanning was conducted during alternations between the experimental condition, during which topographical representations were learned and retrieved, and a control condition, designed to control for primary visual and motor aspects of the task. The control condition was a different environment consisting of an endless, looping corridor—thus containing negligible topographic detail. Subjects were instructed to continually move through the corridor during the control condition. Each image acquisition period consisted of four control and four experimental conditions, each 45 s in duration.

Prior to the imaging experiment, all subjects were trained on a different maze to familiarize them with the task conditions and the operation of the game pad.

**Data Acquisition and Analysis**

**fMRI Scanning**

Following the acquisition of sagittal and axial T1 weighted localizer images (TR = 600 ms, TE = 15 ms), gradient echo, echoplanar fMRI was performed in 24 contiguous 5 mm axial slices (TR = 3000 ms, TE = 50 ms) using a 1.5 T GE sigma system equipped with a prototype fast gradient system and the standard quadrature head coil. Head motion was minimized using foam padding. Each image acquisition session was 6 min in duration, resulting in 120 images per slice and a total of 2880 images per acquisition. In-plane motion correction was performed by shifting images in both transverse directions to minimize the squared difference from the first image in the series.

**Individual Subject Analysis**

Functional data were correlated with a square reference function representing the alternations between experimental and control conditions. The resulting correlation map was filtered to remove pixels with non-physiologic behavior, including those not overlying brain parenchyma or with large intensity signal changes (8%). A nearest-neighbor algorithm removed clusters consisting of less than three contiguous pixels. Maps were thresholded at \( t > 0.2 \). Our prior experience using identical filtering and thresholding on fMRI runs obtained without task activity demonstrated infrequent false positive activation at this level. Suprathreshold correlations were superimposed upon T1 weighted images using a color scale.

**Region of Interest Analyses**

For determination of degree of activation and significance levels, regions were defined for several anatomical areas. Medial-temporal regions (entorhinal/perirhinal cortex, parahippocampal and hippocampus proper) were defined upon axial T1 scans for each subject using a combined MRI/anatomical atlas (Duvernoy, 1991). These regions were then propagated through each fMRI activation run to identify the number of pixels reaching \( t > 0.2 \). This quantification of activation was normalized to the volume of the defined area and paired t-test comparisons were made with control regions defined in the lateral temporal lobes.

**Pseudo-subject Overlay**

Points of activity judged to be significant for each individual subject were mapped to standardized Talairach space (Talairach and Tournoux, 1988) to show the union of all activity observed in individual subjects. No spatial smoothing of functional data was used for this analysis.

**Regression Statistical Parametric Mapping (SPM)**

Data from all subjects were combined using the regression SPM technique described by Friston (1994/1995). Raw time series data from each subject were collapsed to produce eight observations for each voxel, corresponding to the mean value of each 45 s experimental or control epoch. These data were then spatially normalized (Talairach and Tournoux, 1988) and smoothed by convolving with a three-dimensional, 18.75 mm FWHM Gaussian kernel. The eight observations from each subject served as the dependent variable array for regression analysis with the independent variables being task (i.e. alternation between experimental and control conditions), block (i.e. the first and second half of the scan, used to partially correct for linear drift in the signal) and subject. The effects of task were assessed by creating a map of \( t \)-values corresponding to the parameters of task. Assuming that the eight observations from each subject are independent, likely because collapsing the time series data minimizes temporal auto-correlation, this analysis results in a measurement with 59 degrees of freedom.

The \( t \) field result of Worsley (1994) was utilized to derive a critical \( t \)-value for the entire map. Given values for degrees of freedom, smoothness, search volume, desired minimum 'blob' volume (0.5 cm\(^3\)) and desired alpha value (0.02), a critical \( t \)-value of 4.5 was calculated. The SPM map was thresholded at this \( t \)-level and individual 'blobs' below the minimum volume removed. The Talairach coordinates of local \( t \) maxima within the observed regions of activity were determined.

The axial T1 weighted localizer images from each subject were converted to Talairach space and averaged. The resulting anatomical map demonstrates the resolution of the spatial conversion routine and is used for display of the SPM results.

**Results**

All nine subjects required between two and four learning sessions to produce a map of sufficient accuracy. Figure 3 is a reproduction of one subject's final map and demonstrates the level of detail required. Topographic relationships, spatial distances and object locations are all correctly represented. All subjects performed the navigation tasks with a high level of accuracy. Most (6/9) subjects travelled from all four object locations back to the maze start without error. The remaining subjects made infrequent (one or two) wrong turns, but all quickly realized their error and returned to the correct path. Functional data acquired during the first and last learning runs, along with the retrieval task, were analyzed for every subject.

**Single Subject Activity**

The scans for each subject were individually analyzed and thresholded to display task correlated activity. All nine subjects demonstrated activation in the parahippocampal gyrus during the first learning condition. This site of activity was present for eight of the nine during the last learning and six of the nine during retrieval. The location of this activity was determined by anatomical landmarks and conversion to Talairach coordinates. No significant activation was observed in the hippocampal formation, or the entorhinal or perirhinal cortex. Due to susceptibility effects, the fMRI signal was lost from the most anterior and inferior portion of the temporal lobes and thus these areas were not reliably observed. However, the hippocampal formation, over its entire extent, was imaged in all subjects. Region of interest analyses of medial-temporal areas
Figure 4. Individual subject fMRI results. (A) Functional activity in the parahippocampal gyrus during learning and retrieval in three typical subjects. The axial slices roughly correspond to the level indicated in the inset sagittal view. Areas of task correlated activity are shown as colored points over the grayscale anatomical image. The scale shows the colors used to represent pixels which exceed a given r value. (B) Pseudo-subject projection showing combined medial-temporal activity in all subjects during the first learning run. The boundaries of the hippocampal formation, entorhinal cortex, parahippocampal cortex and perirhinal cortex are indicated in blue. The red and yellow points indicate the number of subjects with overlapping activity at a given location and are projected upon orthogonal anatomical atlas slices which are parallel or perpendicular to the bicommissural line and pass through the centroid of the parahippocampal activity.

revealed that only activity in the parahippocampus achieved significance (P = 0.03, 0.02 and 0.03 for first learn, last learn and retrieve respectively) when compared with control regions.

Three of the nine subjects demonstrated bilateral parahippocampal activity at our display thresholds, the remainder showing unilateral activity, evenly divided between the right and the left. Figure 4A shows the medial-temporal activity in three representative subjects during learning and retrieval of topographic information. Region of interest analyses confirmed that activity in the parahippocampus was not significantly lateralized. Figure 4B is a spatially normalized overlay of individual subject activity, illustrating the union of medial-temporal activity observed in all subjects during their first exploration of the maze.

Group Activity
Regression SPMs were generated for the first and last learning runs as well as the retrieval task. No differences between the three conditions were noted at this level of analysis as measured by the extent and anatomical location of activity. Figure 5 is the SPM for the last learning condition. Significant activity across subjects was observed in the right premotor area as well as bilaterally in the superior posterior parietal, posterior cingulate, lingual gyrus and posterior medial-temporal areas. A local maxima analysis was undertaken to reveal the sites of focused activity (Figure 5 and Table 1). Among other areas, foci were present bilaterally in the parahippocampal gyrus.

Effects of Learning
The pattern of activation observed across learning and retrieval was largely constant. Region of interest analyses of the parahippocampus revealed that activation was greater during the learning conditions than during retrieval (P = 0.04). No significant differences in regional distribution of activation were observed in SPMs generated for the first learn, last learn or explore condition.

Discussion
Several cortical areas were robustly activated by maze learning and retrieval. We discuss first the general pattern of cortical
activity observed, including those areas thought to be related to spatial and non-spatial aspects of the task. Activity in the parahippocampus, the only area of activity within the hippocampal system, is then discussed in greater detail.

Spatial Processing
A fundamental feature of spatial information processing is the need to transform the coordinate frame in which stimuli are coded (Anderson et al., 1993). Because visual information, for example, is obtained in retinotopic coordinates, it must be combined with a representation of eye and head position in order to allow responses within the egocentric (body-centered) coordinate frame. This study required subjects to create an allocentric representation of space, encoding spatial locations in terms of their topographic relationship to one another. In order to accomplish this task, subjects presumably combined primary visual stimuli with information regarding their ‘virtual’ body position and with an integrated measure of movement within the environment. Several models of spatial processing have posited a network of regions, including the posterior parietal cortex, retrosplenial cortex, parahippocampus and hippocampus, which participate in the production of an allocentric sense of position in this fashion (see e.g. McNaughton et al., 1991). These proposals are based upon neurophysiological studies which reveal cell populations responsive to stimulus location in retinotopic, head-centered or body-centered coordinates, subject position or heading, and angular velocity. In the present study, activity observed in the parietal and posterior cingulate/retrosplenial cortex with maze exploration and navigation likely reflects recruitment of this spatial processing

<table>
<thead>
<tr>
<th>Anatomical area</th>
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<tr>
<td>1 cerebellum</td>
<td>R 4 -82 -22</td>
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</tr>
<tr>
<td>2 fusiform gyrus</td>
<td>L -37 -67 -14</td>
<td>4.9</td>
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<tr>
<td>3 L -30 -55 -14</td>
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<td>4.8</td>
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<td>4 parahippocampus</td>
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</tr>
<tr>
<td>5 L -15 -52 -3</td>
<td></td>
<td>5.9</td>
</tr>
<tr>
<td>6 posterior cingulate</td>
<td>L -13 -70 -9</td>
<td>6.8</td>
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<tr>
<td>7 R 4 -70 -9</td>
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<td>14 L -33 -52 -39</td>
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<tr>
<td>15 middle frontal gyrus</td>
<td>R 23 -7 -57</td>
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Figure 5. Regression SPM analysis of the last learning scan for all subjects. Significant (t > 4.5, α = 0.02 [corrected for multiple observations]) activity is shown in color superimposed upon averaged axial slices converted to Talairach space. The top row illustrates the approximate location of the entorhinal/perirhinal cortex, parahippocampus and hippocampus. The numbered local maxima correspond to the points listed in Table 1.

Table 1
Local maxima of activity within the SPM presented in Figure 5
representation is not its sole function, this study provides evidence that the parahippocampus plays a crucial role in topographic learning in humans. The use of a virtual environment allowed the examination of topographical learning in intact human subjects confined to the magnet bore, a general approach with several potential applications within the cognitive neurosciences.

Notes
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Address correspondence to Mark D’Esposito, MD, Cognitive Neurology Section, Department of Neurology, Hospital of the University of Pennsylvania, 3400 Spruce Street, Philadelphia, PA 19104-4283, USA.

References