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Spatial Deficits in a Virtual Water Maze in Amnesic Participants with Hippocampal Damage

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ABSTRACT: The Morris water maze is a standard paradigm for the testing of hippocampal function in laboratory animals. Virtual versions of the Morris water maze are now available and can be used to assess spatial learning and memory ability in both healthy and brain injured participants. To evaluate the importance of the hippocampus in spatial learning and memory, we tested five amnesic participants with selective hippocampal damage using a virtual water maze called the Arena Maze. The amnesic participants with hippocampal damage were impaired on the invisible platform (place) task that required them to use distal cues, but were able to navigate almost as well as comparison participants when the invisible platform was marked by a single proximal cue. These results not only confirm that the hippocampus plays a necessary role in human navigation in large-scale environments but also provides a new link between the mnemonic and navigational roles of the hippocampus.

KEY WORDS: hippocampus; amnesia; spatial navigation

INTRODUCTION

Many investigators agree that the hippocampus and adjacent medial temporal lobe structures are essential for declarative memory (Squire et al., 2004; Moscovitch et al., 2005) and memory for the spatial context of events (King et al., 2004; Nadel and Hardt, 2004). The key role of the hippocampus in animal navigation is well known. Tolman (1948) proposed that rats make "cognitive maps" (or spatial representations) for navigation and 30 yrs later O'Keefe and Nadel (1978) described the cognitive map as a representation of the physical environment and one's location within that environment. The discovery of "place cells" in the rat hippocampus (O'Keefe and Dostrovsky, 1971) showed that animal navigation actively engages this structure, and lesion studies confirmed the essential role of the hippocampus in place learning and memory (O'Keefe and Nadel, 1978). Subsequent lesion studies have compared navigation using a cognitive map to navigation using a combination of cues and responses. One task, the Morris water maze (Morris, 1984), has provided a definitive paradigm that shows hippocampal lesions produce a deficit in place learning but not in cue-response learning in rodents (Morris et al., 1982).

A number of brain regions including the hippocampus are necessary for various aspects of spatial memory and navigation. For example, the parahippocampal cortex is thought to be involved in acquiring new spatial memories (Ross, 1980; Habib and Sirigu, 1987; Aguirre et al., 1996; Barrash et al., 2000) and neuroimaging studies find retrosplenial activity during virtual navigation tasks (Maguire et al., 1997; Grön et al., 2000; Mellet et al., 2000; Hartley et al., 2003; Iaria et al., 2003; Rosenbaum et al., 2004; Kumaran and Maguire, 2005; Spiers and Maguire, 2006). Further, the caudate mediates stimulus-response navigation (Packard and McGaugh, 1996; Hartley et al., 2003; Iaria et al., 2003; Bohbot et al., 2004; Voermans et al., 2004; Etchamendy and Bohbot, 2007) and the parietal cortex supports remote spatial memory through egocentric mental representations (Spiers and Maguire, 2007).

Imaging studies using virtual environments find the hippocampus is active during spatial navigation tasks (Maguire et al., 1998; Bohbot et al., 2004; Parslow et al., 2004). Further, impairments in navigational memory in virtual or videotaped environments occur following temporal lobectomy (Maguire et al., 1996; Spiers et al., 2001). Virtual versions of the Morris water maze (MWM) have been developed to assess the role of the hippocampus for spatial navigation and memory in humans using a task analogous to that used for rodents. Impairments in performance in virtual reality versions of the MWM are reported in older individuals (Driscoll et al., 2005), when NMDA receptor antagonists are present (Rowland et al., 2005), and following traumatic brain injury (Skelton et al., 2000, 2006). These place learning deficits in individuals with traumatic brain injury with concomitant hippocampal damage occur in combination with preserved cue-response learning (Livingstone and Skelton, 2007). Moreover, hippocampal and parahippo-

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campal theta oscillations measured in humans during navigation to a hidden platform in a virtual reality MWM found spatial learning is dependent upon normal hippocampal and parahippocampal theta oscillations (Cornwell et al., 2008). Finally, patients with unilateral hippocampal resection have severe spatial memory impairments in a virtual MWM task (Astur et al., 2002). However, there is little information regarding the relationship between focal bilateral hippocampal damage and place learning in a virtual version of the MWM in humans.

To investigate the nature of deficits in place learning after hippocampal damage, we examined the performance of well characterized amnesic participants with damage limited to the hippocampus due to anoxic brain injury. These amnesic participants have been the focus of extensive declarative memory research studies on chronic amnesic syndromes (Hopkins et al., 2004; Wais et al., 2006; Shrager et al., 2007). To be considered amnesic (Squire and Zola, 1997), the amnesic participants in the current study were required to receive a score at least one standard deviation (SD) below the normal range on the General Memory Index of the Wechsler Memory Scale-III (WMS-III), but within the normal range on the Wechsler Abbreviated Scale of Intelligence (WASI) Full Scale Intelligence Quotient (FSIQ) and the WMS-III Working Memory Index. These amnesic participants display significant declarative memory deficits, but have normal intellectual function and spared nondeclarative memory. We, therefore, identified this group as ideal for studying the role of the human hippocampus in spatial learning and memory. We used a virtual reality version of the MWM (Skelton et al., 2006; Livingstone and Skelton, 2007) to test the ability of these amnesic participants to navigate to a platform in several conditions (visible, place, and landmark). The parameters of the virtual reality version of the MWM were similar to the MWM used with rats. On the basis of the prior research, we predicted amnesic participants would be impaired in navigating in the place condition, but would retain the ability to navigate in the visible platform condition. Performance in the landmark condition was less predictable because entorhinal lesions have been shown to spare navigation to proximal stimuli (Parron et al., 2004), whereas hippocampal lesions produce a partial deficit in navigation to proximal stimuli, unless the cue is contiguous with the platform location (Save and Poucet, 2000).

MATERIALS AND METHODS

Participants

Amnesic participants with hippocampal damage and healthy comparison participants matched for age, gender, and education level were included in this study. There were five amnesic participants, four males and one female and five comparison participants, four males and one female. Neither the amnesic nor comparison participants had prior neurological disorders, alcohol or drug abuse, or psychiatric disorders. The amnesic participants, as shown by their performance in other memory studies, have stable nonprogressive cognitive deficits. This study was approved by the Brigham Young University Institutional Review Board and conformed to institutional and federal guidelines for the protection of human subjects. Written informed consent was obtained prior to behavioral testing in all participants. Participants completed a short background information questionnaire detailing age, gender, education level, and brief neurological/psychiatric history.

Magnetic Resonance Imaging

MR images were acquired at General Electric 3.0 Tesla Scanner (GE Medical Systems, Milwaukee, WI) using standard clinical protocols. Sagittal T1-weighted (TR/TE/excitations = 500/11/2) images were acquired and used for localization with a 24 cm field of view. With the midsaggital image as a reference, axial followed by contiguous axial proton density (TR/TE = 2,500/15) and contiguous T2 weighted (TR/TE = 5,253/93.6) spin echo images were acquired, with a slice thickness of 5 mm. Images were acquired on a 256 × 256 matrix with a 22 cm field of view for the axial images. Contiguous T1 coronal images were acquired, (TR/TE = 13/4.47) 1.2 mm thick followed by coronal contiguous T2 weighted images (TR/TE = 3,500/114) of 1.5 mm thick. Coronal images were acquired with a field of view of 25.6 cm on a 256 × 256 matrix. A neuroradiologist rated all scans for gross lesions or other abnormalities.

Volumetric Image Analysis

Proton density and T2 axial dual-echo images were quantified as described by Blatter et al. (1995) using the software ANALYZE 5.0/6.0® (Biomedical Imaging Resource, Mayo Foundation, Rochester, MN). The original 16-bit images were converted to 8-bit images in ANALYZE® file format. A multistep volume analysis was then performed using several image-processing tools available in ANALYZE. Regions of cerebral spinal fluid (CSF), white matter and gray matter were defined by the user, and plotted in a two-dimensional feature space. Quantitative MR analyses of the temporal lobe gyri were performed as per the methods described previously (Bigler et al., 1997, 2002).

Quantitative MR analyses of brain structures were performed on all participants as per the methods described previously (Bigler et al., 1997, 2002). Volumes of the following brain structures were determined by using the region of interest (ROI) feature of ANALYZE® that yields a count of gray matter, white matter, and CSF: lateral ventricles, third ventricle, fourth ventricle, temporal horns, total brain volume, and cerebral spinal fluid (CSF).

Volumes of the following brain structures were determined by using the ROI feature: hippocampus, parahippocampal gyrus, fusiform gyrus, inferior temporal gyrus, middle temporal gyrus, and superior temporal gyrus. The volumes of the temporal horn of the lateral ventricle, rinal sulcus, inferior temporal

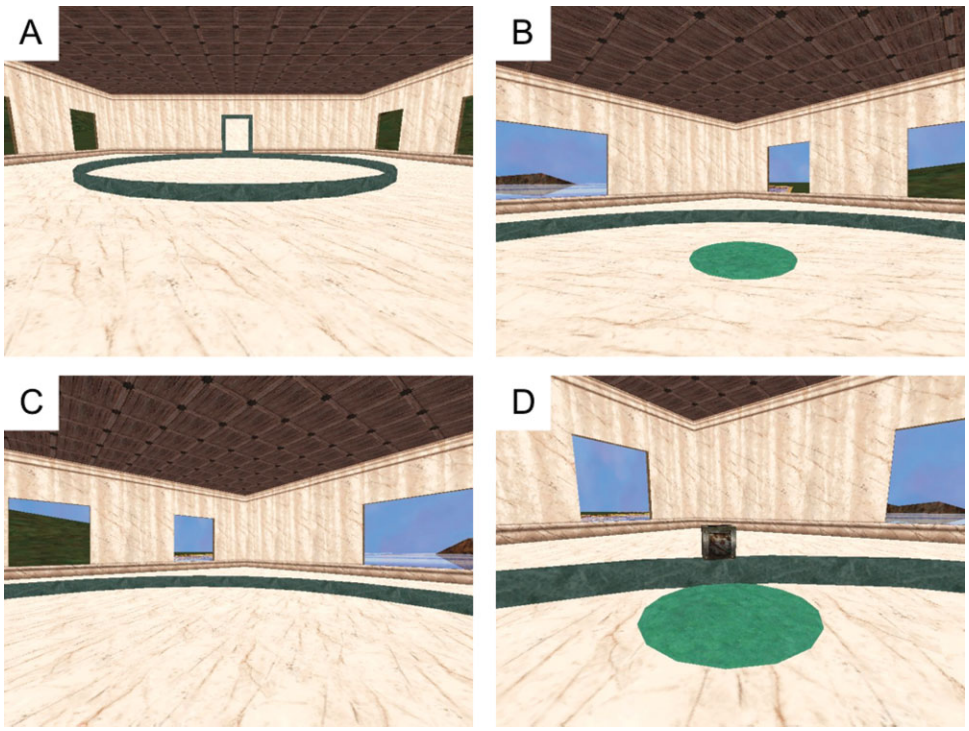

FIGURE 1. Views of arena maze and landmark maze. (A) View from the starting position in the exploration trial, showing the north wall. (B) Visible trial 4 showing the platform visible in the southwest quadrant of the arena. (C) Place trial 1 showing the southeast quadrant. (D) Landmark trial 1 showing the cue object (metal box) on the wall of the arena in the southeast quadrant. The platform location is illustrated; however the platform remains hidden until the participant steps on it. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

lobe sulcus, and sylvian fissure were quantified as well. Temporal lobe volumes encompassed portions of Brodmann areas 20, 21, 22, 25, 27, 28, 34, 36, 37, 38, 41, and 42. For gyral volumes the total number of gray matter and white matter pixels within the ROI for each section and multiplying by the voxel dimensions; the CFS pixels were used to determine sulcal and temporal horn volumes. Tracing was done in the coronal plane and all three planes were used to cross check anatomical markers. We followed a previously published protocol for the temporal lobes (Bigler et al., 2002). Intrarater and interrater reliability exceeded 0.90.

Hippocampal volumes were measured in the coronal slices (Bigler et al., 1997). The hippocampal formation was manually traced in a posterior to anterior direction. The starting slice was identified using the following anatomical landmarks: (1) good separation of the lateral ventricles, (2) the appearance of the pulvinar of the thalamus, and (3) the appearance of the corpora quadrigemina. Measurement of the hippocampal formation was discontinued when the temporal horn of the lateral ventricle extended more than half way across the width of the hippocampus. Intrarater and interrater reliability exceeded 0.90.

Neuropsychological Tests

Amnesic patients were administered neuropsychological tests to assess memory, general intellectual ability, and visual-motor function. Memory function was assessed with the Wechsler Memory Scale-III (Wechsler, 1999) and intellectual function was assessed with the Wechsler Abbreviated Scale of Intelligence (Wechsler, 1997). Visual-motor function and spatial memory was also assessed with the Rey-Osterrieth Complex Figure Test (Meyers and Meyers, 1995).

Behavioral Apparatus

The apparatus consisted mainly of a virtual MWM called the "Arena Maze," constructed using the editor supplied with Unreal® (Epic Megagames; Livingstone and Skelton, 2007). The Arena Maze was presented using a laptop computer with a 15-inch monitor set at 800 × 600 resolution. Participants experienced a first person view of a circular arena set in a square room with windows to an outdoor environment. The arena was 40-m in diameter bound by a featureless, 1-m high wall, so that participants could not exit the arena but could easily view the virtual room (75 m × 75 m) and outdoor world (Fig. 1). The walls of the virtual room were distinctive and arbitrarily designated as North (N), East (E), West (W), and South (S). The North wall was featureless except for a door and the South wall had a single large window through which a body of water, a beach, and an island could be seen. The East and West walls each had three windows displaying a mountainous landscape sloping towards the beach. Trials always started at one of the four cardinal points of the arena, that is,

closest to one of the four walls. Participants moved around the virtual environment using a preprogrammed game pad that allowed forward (5 m/s) and left or right (30 degrees/s) movements. Backward movements were not permitted, to simulate the "real world" movement of swimming rats performing the MWM. Participants were further embedded into the environment in that they heard sounds of footsteps and experienced a slight "head bob" when walking.

Arena maze task

The Arena Maze task consisted of four types of trials presented in the following sequence: one exploration trial, four visible platform trials, 10 invisible platform place trials, and one probe trial. During the exploration trial, participants were exposed to the features of the environment and trained to use the game pad. At a minimum, participants approached all of the windows and looked at the landscape outside of the room. During the four visible platform trials, a large (7 m) circular platform was visible on the floor of the arena and its location varied on each trial. These trials tested the ability of participants to become familiar with task demands (i.e., to move quickly and directly to the platform) and to navigate to a visible target in the environment. During the 10 place trials, the large (7 m) platform was always located in the Southeast quadrant of the arena and was invisible until stepped on at which point it would rise slightly above the floor and make a mechanical sound to alert participants that they had reached the target location. During the probe trial, no platform was present, but after 50 s the missing platform would automatically rise and make the usual mechanical sound whether or not a participant had stepped on it. The place trials tested the ability of participants to find the platform using only spatial cues and the probe trial tested the place learning of the target location. The start position of the exploration trial was in front of the single window on the South wall of the room. The start positions for all remaining trials were just inside the wall of the arena at one of the cardinal points and varied in a fixed, pseudorandom order. The trial start position sequence for the place and probe trials were: WENS, ENWS, NS, and W.

Landmark maze

The Landmark Maze tested the ability to associate a hidden target location with a single landmark object (i.e., stimulus-response navigation). In this variation of the Arena Maze, there were five invisible platform trials all starting from the North start position. The platform was located near the edge of the arena (i.e., adjacent to the arena wall) and in a different position on each trial. The platform location for each trial was: SE, SW, E, W, and SE. A single landmark object (a gray metal box) was positioned on the arena wall close to the invisible platform to indicate its location.

Behavioral Procedures

Maze testing sessions were conducted with the participant seated in front of the computer and the experimenter behind the participant and out of view. Before testing began, participants were told that they would be moving through several virtual rooms to complete a number of tasks and that their main goal was to locate a green platform on the floor. Next, participants were given the game pad and allowed to explore the virtual room for as long as they liked (exploration trial). At a minimum, participants approached all of the windows and looked at the landscape outside of the room. Experimental trials began when participants indicated they were finished exploring and when the experimenter was sure they were comfortable with the game pad.

Participants began with the four visible platform trials of the Arena Maze. After completing the visible platform trials, participants were informed that the target platform would now be invisible, but would always be in the same place. They were asked to go to the platform as quickly and directly as possible. They were also told that on one of the trials the platform would be "very difficult to find" (probe trial). After each place trial, participants were prompted to look about the room, while remaining on the platform. This indirectly encouraged participants to learn the spatial layout of the virtual environment and location of the invisible platform. For all place trials, if participants failed to find the target within 3 min, they were given verbal directions to get to the invisible target (e.g., "turn left," "go straight"). As stated previously, the probe trial (i.e., the "very difficult to find" trial) always came after the 10 place trials. After completion of the Arena Maze trials, participants were given instructions for the five landmark trials. They were told that the platform would be in a different place on each trial, but they should still try to go to the platform as quickly and directly as possible.

After completing all of the maze trials, participants completed the Room Reconstruction task to assess the degree to which they had learned the spatial layout of the room and associated the platform with the correct location in the layout. Participants arranged laminated pictures of the four walls around a picture of the arena floor according to their understanding of the spatial layout. They were then asked to place a laminated picture of the platform (to scale) on the floor and in the location where it had appeared in the Arena Maze invisible trials. Responses were scored on a scale of 0 to 4: 1 point for correctly placing the two walls nearest the platform, 2 points for adding the correct position of the remaining walls, 3 points for adding the platform in the correct hemisphere, and 4 points for placing the platform in the correct quadrant. A score of zero was given if the two walls nearest the platform were incorrectly positioned.

Analysis of Behavioral Measures

Behavioral data from testing in the mazes were extracted from "Demo files" recorded during navigation and analyzed

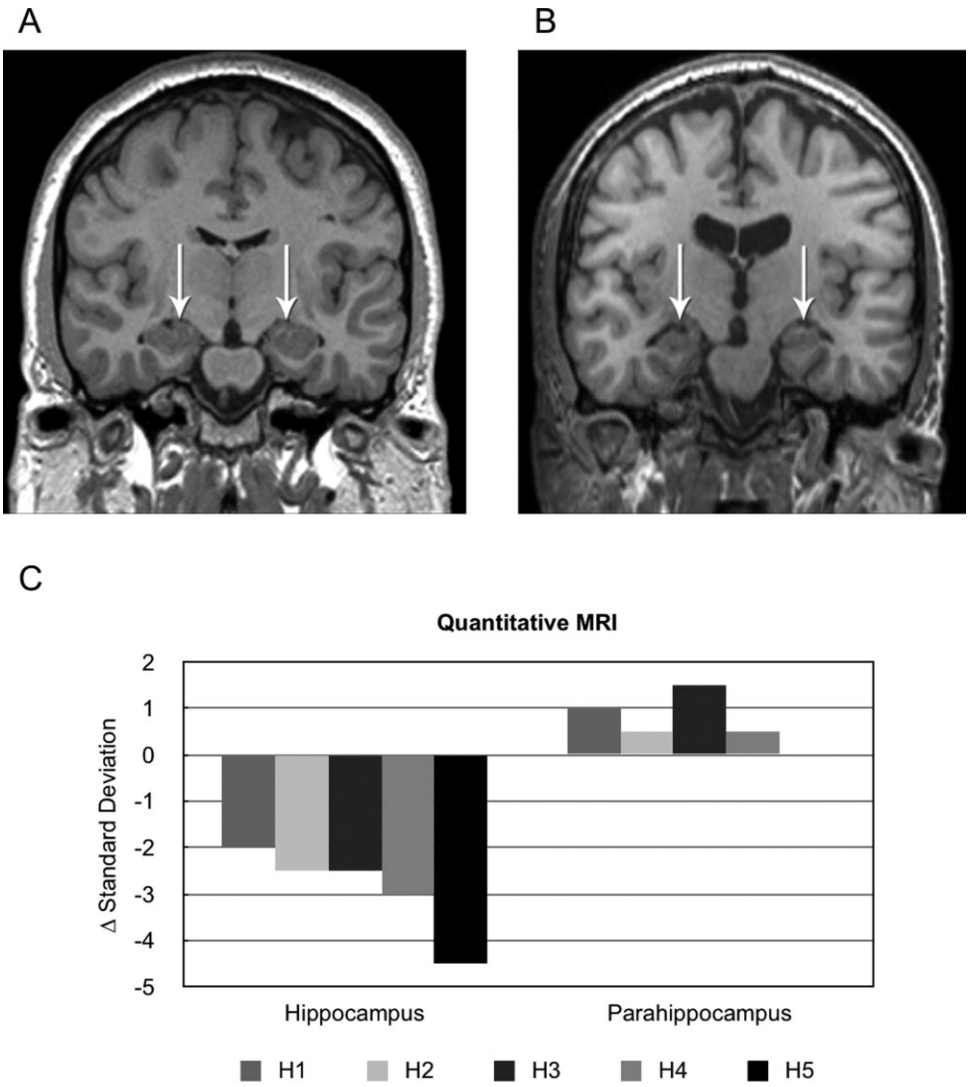

FIGURE 2. Clinical brain MRI reports by a neuroradiologist for the amnesic participants indicated no evidence of lesions or other structural abnormalities other than within the hippocampus. Shown are brain scans from (A) a healthy comparison and (B) an amnesic participant with bilateral hippocampal damage. Amnesic participants had hippocampal volumes reduced by an average of 20%. (C) For each amnesic participant, graphed is the number of standard deviations away from the MRI comparison participants' hippocampus and parahippocampal gyrus means. H5 parahippocampal volume was 0 standard deviations from the MRI comparison participant and therefore does not appear on the graph.

using TRAM® software (Skelton et al., 2006). Data were analyzed using conventional measures adopted from the MWM paradigm including latency (time to reach the platform) and distance (distance traveled to the platform). For the probe trial, we calculated the percentage of time spent searching in the correct quadrant (where the platform had been located) and the other three quadrants. Statistical analyses were conducted using Microsoft Office Excel and SPSS®. Individual repeated measures ANOVAs with trial as the repeated measure were conducted separately for latency and distance for each of the three maze conditions: visible, place, and landmark trials. Because sample sizes were very small and the distribution of data was positively skewed, a logarithmic transformation was used for latency (in s) and distance (in pool diameters). Mann–Whitney *U* tests were used to assess the results of the Room Reconstruction task and *t*-tests were used for all other analyses.

RESULTS

Participants

The mean age of the amnesic participants was 44.25 ± 5.17 yrs (range 30–54 yrs) with a mean educational level of 12.75 ± 0.25 yrs (range 12–14 yrs). The mean age of the comparison participants was 38.5 ± 6.34 yrs (range 24–60 yrs; $t = 0.16$, $P = 0.88$) with a mean educational level 13.0 ± 0.52 yrs (range 12–13 yrs; $t = 0.05$, $P = 0.94$).

TABLE 1.

Average (±S.D.) Neuropsychology Test Results for the Comparison and Amnesic Participants

Neuropsychology test and subtest	Comparison Mean ± S.D.	Amnesic Mean ± S.D.	*P*
WASI			
Full Scale IQ	110.4 ± 10.5	102.4 ± 5.3	0.17
Verbal IQ	105.2 ± 8.0	100.4 ± 7.0	0.34
Performance IQ	112.8 ± 16.1	103.2 ± 12.0	0.32
WMS-III			
Immediate Memory Index	99.8 ± 16.3	76.2 ± 20.3[a]	0.08
General Memory Index	113.0 ± 14.2	84.0 ± 23.5[a]	0.05
Working Memory Index	112.0 ± 10.5	93.8 ± 9.9	0.02
ROCFT			
Copy	n/a	33.6 ± 2.4	n/a
Immediate recall	n/a	11.5 ± 7.6[a]	n/a
Delayed recall	n/a	11.4 ± 8.0[a]	n/a

[a]Scores are more than one standard deviation below the standardized mean. WASI, Wechsler Abbreviated Scale of Intelligence; WMS-III, Wechsler Memory Scale-III; ROCFT, Rey-Osterrieth Complex Figure; n/a, not available.

Quantitative MRI

Clinical brain MRI reports by a neuroradiologist for the amnesic participants indicated no evidence of lesions or other structural abnormalities other than within the hippocampus (see Fig. 2). On the basis of quantitative MR image analyses, all five of the amnesic participants had significant hippocampal atrophy as the right (mean = 1.93 ± 0.20 cm^3) and left (mean = 1.96 ± 0.24 cm^3) hippocampal volumes were more than one standard deviation below the normal comparison group (right hippocampus mean = 2.55 ± 0.13 cm^3, and left hippocampus mean = 2.48 ± 0.13 cm^3). The amnesic participants had significant hippocampal atrophy compared to normal MRI comparison participants for the right hippocampus (t = 6.47, $P < 0.0001$) and for the left hippocampus (t = 4.79, $P < 0.001$), respectively. On average, amnesic participants had 20.4% ± 8.2% right and 19.0% ± 10.1% left hippocampal atrophy compared to normal MRI comparison participants. There were no differences in any temporal lobe gyri volumes for the amnesic participants compared to comparison participants.

Neuropsychological Tests

The results of the neuropsychological tests are reported in Table 1. The amnesic participants' Verbal Intelligence Quotient (VIQ) and Performance Intelligence Quotient (PIQ), and Full Scale Intelligence Quotient (FSIQ) were within average range of intelligence. As expected, the amnesic participants' were significantly impaired on the Immediate and General Memory Index, but not on the Working Memory Index. The amnesic participants' visual-motor function on the copy portion of the Rey-Osterrieth Complex Figure Test was within normal range, but their spatial memory (delayed recall of a figure) was significantly impaired.

Behavioral Measures

Arena maze

The results of the Arena Maze are reported in Table 2. The amnesic participants were able to navigate to a visible platform in the arena as quickly and directly as the comparison participants (Fig. 3). Both groups' performance on the visible platform trials (trials 1–4) demonstrated that they were equally capable of coping with the procedural demands of the task, that is, they were able to use the game controller and follow the task instructions. There was no group effect (comparison and amnesic participants) for latency ($F(1, 8) = 0.92$, $P = 0.37$) or distance ($F(1, 8) = 1.01$, $P = 0.35$) to travel to the visible platform. The trials by group interactions were not significant.

For performance on the place trials (trials 2–10), there was a significant Group effect for both latency ($F(1,8) = 7.17$, $P = 0.03$) and distance ($F(1,8) = 9.38$, $P = 0.02$). Specifically, the amnesic participants were impaired in their ability to locate the platform and took more time and longer paths to reach it than did comparison participants (Fig. 3). The trials by group interactions were not significant.

Performance on the probe trials confirmed the inability of amnesic participants to identify the location of the invisible target location relative to spatial cues in the virtual environment (Fig. 4). Amnesic participants spent significantly less time searching in the correct quadrant (the quadrant that contained the platform during the place trials) than did the comparison participants (amnesic mean = 41% ± 9%, comparison mean = 73% ± 6%, t = 2.51, P = 0.04, d = 1.84). Figure 4A shows the mean percentage latency (dwell time) spent in all four quadrants of the arena for both the amnesic and comparison groups. The notable group difference is further illustrated

TABLE 2.

Statistical Results for the Virtual Morris Water Maze

	Comparison Mean ± S.E.M.	Amnesic Mean ± S.E.M.	*P*
Visible trials (1–4)			
Latency (s)	3.66 ± 0.54	4.58 ± 0.67	0.37
Distance (pool diameters)	0.62 ± 0.02	0.64 ± 0.03	0.35
Place trials (2–10)			
Latency (s)	9.08 ± 1.75	18.81 ± 2.53	0.03
Distance (pool diameters)	0.93 ± 0.09	2.99 ± 0.83	0.02
Probe trial (1) (% dwell time)			
Correct quadrant	73% ± 6%	41% ± 9%	0.03
Spatial score (*z*-scores)	0.00 ± 0.30	−4.67 ± 1.39	0.008
Landmark trials (2–5)			
Latency (s)	5.16 ± 1.24	8.19 ± 1.79	0.15
Distance (pool diameters)	1.14 ± 0.19	1.96 ± 0.75	0.34
Room reconstruction	4 ± 0	0 ± 0	0.008

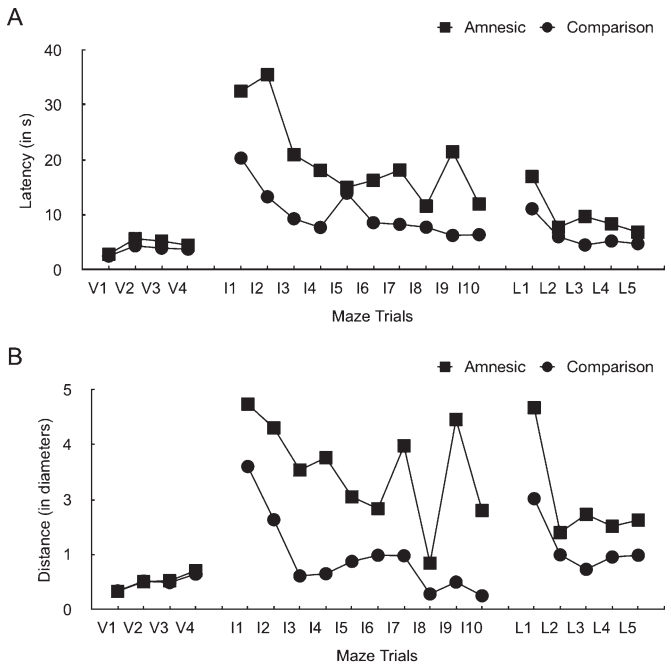

FIGURE 3. Performance on arena maze trials (group means). (A) latency to platform and (B) distance to platform, expressed as a ratio of the diameter of the arena. V, visible; I, invisible; L, landmark.

by the sample paths of one comparison participant and one amnesic participant (Figs. 4B,C, respectively). The search pattern of the amnesic participants was more distributed over the entire arena, whereas the search pattern of the comparison participants was focused largely in the correct quadrant.

The differences between the performances of the groups in the Arena Maze were also compared by calculating a "spatial score" for each participant; the score incorporates latency, distance, and probe dwell time: spatial score = (0.5 × probe *z*-score) − (0.25 × latency *z*-score + 0.25 × path length *z*-score). This omnibus spatial score takes all influences of navigational performance into account (e.g., hesitancy, directness of route, and certainty of location). The scores of amnesic participants were significantly lower than those of comparison participants on this omnibus measure of spatial performance (amnesic mean = -4.67 ± 1.39, comparison mean = 0 ± 0.30, $t = 2.72$, $P < 0.026$, $d = 1.44$). Performance of the groups in the Arena Maze was also compared using a Mann–Whitney *U* test. There was no significant difference between groups on the visible platform trials, but there was a significant difference on the place trials for both latency and distance (data not shown).

Landmark maze

The results of the Landmark Maze are reported in Table 2. The amnesic participants were able to navigate to an invisible platform marked by a single cue almost as quickly and directly as the comparison participants (Fig. 3). When the platform location was marked by a nearby cue for the landmark platform trials (trials 2–5), there were no significant group differences in performance for either latency ($F(1, 8) = 2.59$, $P = 0.15$) or distance ($F(1, 8) = 1.02$, $P = 0.34$). The trials by group interactions were not significant. Performance of the groups in the Landmark Maze was also compared using a Mann–Whitney *U* test. There was no significant difference between groups for latency or distance (data not shown).

Both the amnesic and comparison participants were able to navigate to a platform marked by a single cue. The slight improvement in performance of the amnesic participants on the landmark platform trials over the place trials was reflected in a significant difference in latency ($t(4) = 3.91$, $P < 0.02$), such that performance was better on the landmark trials. However, the amnesic participants showed little improvement in distance compared to their performance in the place condition ($t(4) = 0.68$, $P = 0.53$). Comparison participants had slightly longer paths on the landmark trials than in the place trials, but this difference was not significant. The significant difference in latency, but not distance suggests that the amnesic participants were impaired to a lesser extent in the landmark condition than in the place condition.

Room reconstruction task

Amnesic participants demonstrated poor knowledge of the spatial layout of the virtual room and of the location of the platform. When tested immediately after completing the maze, amnesic participants could not position the four walls of the room in the correct configuration and were unable to indicate where the platform was located. All five amnesic participants received a score of "0" on this task, whereas all comparison participants received a perfect score of "4." Results from a Mann–Whitney *U* test showed that these differences were significant ($U = 0$, $P < 0.008$, $Z = 3.00$, $r = 0.95$).

DISCUSSION

In this study, amnesic participants with focal, bilateral hippocampal damage had significant spatial memory deficits in a virtual water maze, displaying difficulty remembering new spatial locations. The amnesic participants were impaired in navigating to the invisible platform location in the Arena Maze, where there was no nearby landmark available. The amnesic participants also spent more time searching for the platform and took longer paths to reach the platform than did the comparison participants. The impairment occurred even though they were instructed to look around the room after each trial while remaining on the platform. These instructions should have encouraged them to learn the spatial layout of the virtual environment and location of the invisible platform. Further, the place learning deficit was also present in the Room Reconstruction task, where amnesic participants were unable to reconstruct the basic layout of the environment.

Amnesic participants were not impaired at navigating when the platform was visible. On the visible platform trials, the performance of the amnesic and comparison groups was similar. These findings suggest that amnesic participants were able to

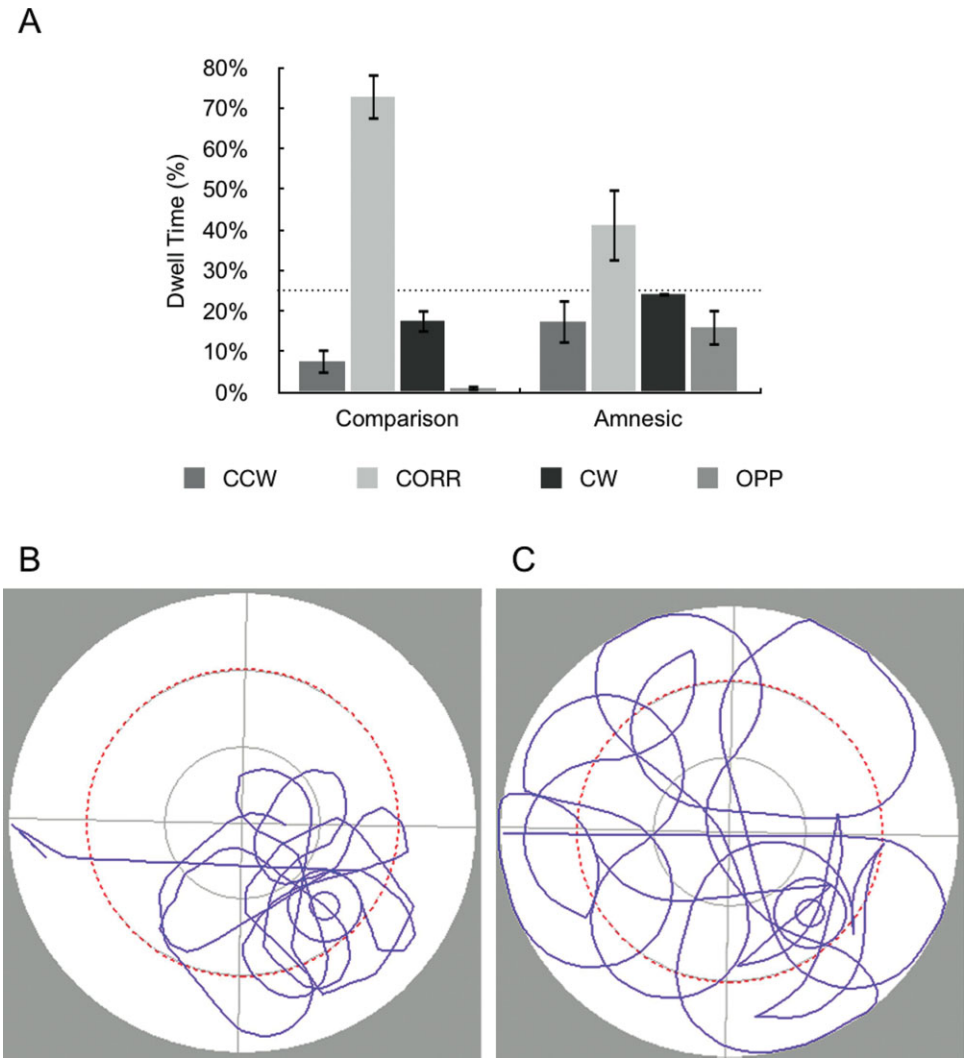

FIGURE 4. Probe trials. (A) Mean + standard error of mean (SEM) dwell times in each of the four quadrants: Correct (CORR), opposite (OPP), and counterclockwise (CCW) and clockwise (CW) from the correct quadrant. The dotted line indicates 25%, or random, responding. (B) Sample paths from the probe trial of a comparison participant who spent 68% of the time in the correct quadrant (i.e., closest to the mean value of the group) and (C) sample path from the probe trial of an amnesic participant who spent 21% of the time in the correct quadrant (i.e., closest to the mean value for the group). [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

navigate to the visible platform using procedurally based navigational strategies, and that they had no motivational, motor, or perceptual problems with the virtual environment task (Dolleman-van der Weel et al., 2009). These findings are consistent with studies on rats with hippocampal damage (Morris et al., 1982) and functional magnetic resonance imaging (fMRI) studies indicating that that the hippocampus is not involved in procedurally based navigation (Hartley et al., 2003; Rauchs et al., 2008).

Amnesic participants were able to find the invisible platform when it was marked by a single proximal cue in the landmark condition almost as quickly and directly as the comparison participants. These results are similar to the previous observation that survivors of traumatic brain injury are significantly better at navigating to an invisible platform when its location is marked by a single cue object (Livingstone and Skelton, 2007). Amnesic participants were slightly better at finding the invisible platform when it was marked by a cue in the landmark condition than when no cue was available during the place trials. The latencies of the amnesic participants were significantly faster, but the distances were not statistically shorter under the landmark condition when compared to the place condition. Studies with hippocampal lesioned rats support our findings. Save and Poucet (2000) showed that rats with dorsal hippocampal lesions were "impaired in the distal and to a less extent in the proximal landmark condition." Rats navigated to a hidden platform based upon the configuration of three proximal cues. Dorsal hippocampal lesioned rats had longer latencies and distances but comparable dwell times on probe trials compared to sham-operated rats. Save and Poucet (2000) did not directly compare performance to distal and proximal landmarks. Dorsal

hippocampal lesioned rats were also not impaired when the cue ("beacon") was at the location of the platform. In a comparable study with the same paradigm, rats with entorhinal lesions were not impaired on the 3-cue proximal landmark condition, although again, performance on the distal and proximal cue conditions was not directly compared (Parron et al., 2004). Thus in our study and these rat studies, direct hippocampal damage produces a preferential but not a completely selective deficit in using distal cues to navigate to an invisible platform.

In the previous study examining the performance of brain injured participants in the landmark condition (Livingstone and Skelton, 2007), the brain injured participants were as good as comparison participants by the end of the landmark condition. In contrast, our amnesic participants were not as good as comparison participants. The discrepancy between these two studies may simply be due to the small sample size of this study. The sample size was small because participants with discrete focal hippocampal lesions are rare whereas participants with traumatic brain injury are, unfortunately, relatively common in the community (Livingstone and Skelton, 2007). Alternatively, the greater impairment in the amnesic participants could have resulted from more substantial hippocampal damage in our amnesic population than that observed in the traumatic brain injury population. The hippocampal damage could have prevented the amnesic participants from perceiving, learning or remembering the association between the cue object and an invisible goal location that varied from trial to trial.

Given that the hippocampus is thought to be necessary for spatial navigation, the present results show that hippocampal loss preferentially disturbs spatial navigation based upon map-like representations. Our findings are consistent with place learning deficits in virtual space shown by humans after temporal lobectomy (Astur et al., 2002) and extend these results through the examination of more focal lesions. Our findings are also consistent with virtual navigation tasks in which hippocampal activity correlates with spatial navigation (Hartley et al., 2003).

The amnesic participants were impaired on the Room Reconstruction Task. Thus, immediately after testing in the virtual environment, participants with focal hippocampal lesions were unable to reconstruct the virtual environment. In other words, amnesic participants were unable to recall or reconstruct elements of the Arena Maze that would constitute a map-like representation of the virtual environment (Jacobs et al., 1998), even with only a very brief delay. These results are consistent with the finding that hippocampal damage impairs the recall of object locations within a virtual environment when there was a shift in viewpoint (King et al., 2004; Shrager et al., 2007) and with the finding that hippocampal damage impairs reconstruction of the arrangement of 16 objects (Smith and Milner, 1981). Although this initial study does not discern whether the deficit seen here was in the ability to perceive or encode the configuration of distal cues, or to retrieve the configuration to use it to navigate in the virtual environment, these present results provide new support for the link between the mnemonic and navigational roles of the human hippocampus.

Our results provide further support for analogous spatial memory processes of the hippocampus in humans and rats. Thus, amnesic participants with selective hippocampal damage performed similar to hippocampal-damaged rodents on the visible and invisible platform trials of the MWM (Morris et al., 1982). Similarly, our amnesic participants, like rats with dorsal hippocampal damage (Save and Poucet, 2000), showed a reduced impairment in navigation to an invisible platform marked by a single proximal cue. It has been difficult to determine homologies between human and nonhuman mnemonic mechanisms for spatial information processing, because methodologies and neuropathology differ (Kesner and Hopkins, 2006). However, recent review (Kesner and Hopkins, 2006) found the rat and human hippocampus subserve similar functions by comparing data across analogous tasks. In particular, similar deficits are observed in rats and humans with hippocampal damage on analogous tasks testing short-term and immediate-term memory, spatial and temporal pattern separation, pattern association, pattern completion, and sequential learning. Our findings provide further evidence that research with laboratory animals can lead to development of novel approaches to better characterize the neuropathology and behavioral performance in humans.

This study assessed the performance of amnesic participants with damage limited to the hippocampus and found spatial memory deficits in humans that are similar to those observed in rodents with hippocampal damage using analogous spatial memory tasks (real and virtual MWM). These findings also suggest that that the human hippocampus is an important (and perhaps necessary) structure for learning to find one's way in a large-scale environment. These data provide support for evolutionary continuity in cognitive function assigned to the hippocampus of rats and humans.

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