Brain regions associated with acquisition and retrieval of verbal episodic memory

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It is widely held that conscious recall of past experiences involves a specific system—episodic memory[1]. Patients with amnesia have gross impairments of episodic memory while other kinds of memory remain intact[2,3], suggesting that a separable brain system underlies episodic memory. We have used positron emission tomography (PET) to identify components of this system in normal volunteers. A dual-task interference paradigm[4] was used to isolate brain areas associated with acquisition, and a cueing paradigm[5] to isolate the areas concerned with retrieval from verbal episodic memory. Acquisition was associated with activity in the left prefrontal cortex and the retrosplenial area, whereas retrieval was associated with activity in right prefrontal cortex and the precuneus. Our results provide clear evidence that episodic memory involves a network of specific prefrontal and posterior structures[6,7] which can be fractionated into different component processes.

Brain activity was measured in six volunteers while they learned and, in another six, while they retrieved verbal material. All volunteers heard 15 paired associates, consisting of categories and relatively uncommon exemplars (such as furniture–sideboard) and were told they would have to recall them later. Recall after only one presentation was ~80% correct so that scanning during one occurrence of a list was sufficient to capture the brain activity associated with consistently successful encoding or retrieval. During the acquisition phase, presentation of words automatically leaves legacies (priming[1]) of the semantic memory[1] processes involved. These can be separated from the processes underlying episodic memory[6,8]. Our experiments were designed to isolate the two types of encoding by subtracting a condition involving only the former processes from one involving both types. Episodic memory encoding is impaired by a difficult but structurally unrelated distractor task[4] while automatic priming is unaffected[8,9]. Experiment 1 (Table 1) contrasted

TABLE 1 Description of tasks

	Paired-associate task	Memory performance	Distractor task*
Experiment 1 (acquisition)			
I	Encoding†	83 ± 4%	Easy
II (control)	Passive listening‡	—	Easy
III	Encoding†	68 ± 4%§	Difficult
IV (control)	Passive listening‡	—	Difficult
Experiment 2 (retrieval)			
	Paired-associate task	Memory performance	
I	Recall‖	81 ± 5%	
II	Generation¶	—	
III (control)	Repetition#	—	

* A joy-stick was used to move a cursor into a box in one of four different positions on a screen in a completely predictable (easy condition) or random (difficult condition) sequence. The inter-trial interval was 1 s.

† Volunteers tried to remember 15 paired associates (for example, poet–Browning) which they heard at the rate of 1 per 3 s during the scan. Immediately after the scan, volunteers filled in a mood questionnaire to prevent rehearsal of the word pairs. Recall was tested 5 min after the end of the scan.

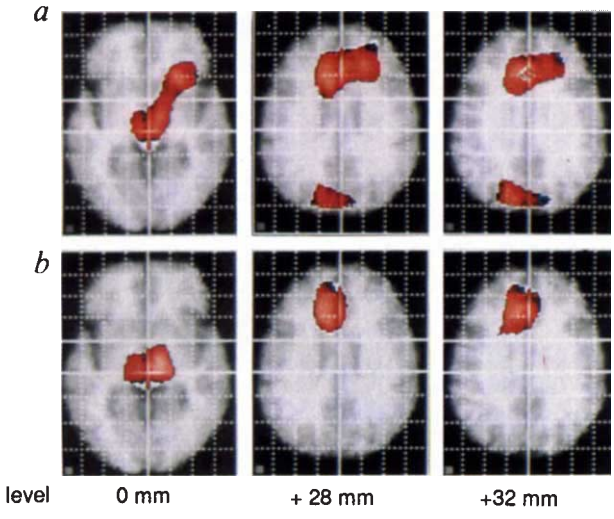
‡ Volunteers heard the same pair of words (one thousand–two thousand) 15 times at the same rate.

§ Significantly inferior to performance with easy distraction: related-t (d.f. = 5) = 7.3, $P < 0.001$.

‖ 15 paired-associates were presented, exactly as in experiment 1, 5 min before the scan. During the scan, volunteers were presented with category labels (for example, poet) at a rate of 1 per 3 s, and had to supply the appropriate, specific exemplar (such as Browning, or the word 'pass').

¶ As in the recall condition, except that new categories were used that had not been presented previously, and the instructions were to generate any appropriate exemplar.

Subjects were presented with a (different) word to repeat every 3 s.



◀ FIG. 1 Regions significantly activated in the memory encoding condition (compared with passive listening) while performing *a*, the easy distracting task and *b*, the difficult distracting task. Areas of significant activation ($P < 0.001$) have been superimposed onto averaged magnetic resonance imaging scans (from 6 normal subjects) which have been transformed stereotactically to fit a standard atlas[25]. Three transverse planes are shown. Levels are relative to the intercommissural plane. In both comparisons there is activation of the left superior temporal gyrus and of the left anterior cingulate cortex. Activation of these areas would be expected given the auditory word processing and attentional demands of the tasks[26,27]. The left medial frontal gyrus (not visible in these slices; Table 2), activated primarily during difficult distraction, may be associated with the additional processes involved in performing two tasks at the same time. Left dorsolateral prefrontal cortex and retrosplenial cortex were activated only during easy distraction. A total of 12 right-handed male volunteers (6 in each experiment), who gave written informed consent, took part in the study, which was approved by the local hospital ethics committee and ARSAC (UK). Scans were obtained using a CTI model 953 B-PET scanner (CTI I, Knoxville, USA) with collimating septa retracted. Volunteers received a 20-s intravenous bolus of $H_2{}^{15}O$ at a concentration of 55 MBq ml^{-1} and a flow rate of 10 ml min^{-1} through a forearm cannula. Images were examined in ANALYZE (BRU, Mayo Foundation, Rochester, MN, USA)[28]. Statistical analysis was performed in PROMATLAB (Math Works, Natick, MA, USA) using statistical parametric mapping (MRC Cyclotron Unit). The original brain images were transformed[29] into a standard stereotactic anatomical space[25]. Resultant images extended from −20 mm below to 60 mm above the intercommissural plane. Global differences in cerebral blood flow were covaried out and significant differences between conditions were then estimated on a pixel-by-pixel basis using the t statistic[30].

TABLE 2 Activation foci in the different conditions

Acquisition (experiment 1)	Easy distractor				Difficult distractor			
	x (mm)	y (mm)	z (mm)	Z value	x (mm)	y (mm)	z (mm)	Z value
L. sup. temp. (22)	−56	0	−4	5.0	−54	−6	0	5.2
R. sup. temp. (21)	—	—	—	—	48	4	−8	4.1
L. ant. cing. (32)	−2	28	28	4.0	−4	22	28	5.4
Med. frontal (9/10)	—	—	—	—	−22	36	20	4.0
L. Prefrontal (46)*	−31	34	8	4.1	—	—	—	—
Retrosplenial (31/23)*	−2	−62	12	4.1	—	—	—	—
Retrieval (experiment 2)	**Episodic**				**Semantic**			
	x (mm)	y (mm)	z (mm)	Z value	x (mm)	y (mm)	z (mm)	Z value
L. ant. cing. (32)	−2	18	36	9.4	−2	20	36	6.9
R. thalamus	2	−22	0	7.5	6	−20	0	6.4
L. thalamus	−2	−22	8	5.1	−8	−24	12	4.1
R. frontal (47)*	26	18	0	5.5	—	—	—	—
R. frontal (10/46)*	18	28	24	4.4	—	—	—	—
L. precuneus (31)*	−6	−68	36	6.1	—	—	—	—
R. precuneus (31)*	12	−72	28	4.0	—	—	—	—

In both experiments 12 PET scans were collected and divided into blocks of 4 or 3 in which each condition was represented. To emphasize episodic memory as opposed to priming, low-frequency category-exemplar pairs were used and explicit memory instructions given in conditions 1.I, 1.III and 2.I of Table 1[2,6]. Coordinates refer to the location, in the stereotactic space defined in ref. 25, of the maximal activity indicated by the highest Z score in a particular cerebral structure. Numbers in parentheses refer to Brodmann areas which are given to help localization of activity rather than to imply any cytoarchitectonic correlates.

* Areas where direct comparison revealed significant differences between the two conditions. Abbreviations: L, left; R, right; sup., superior; temp., temporal; ant., anterior; cing., cingulate; med., medial.

the effects of easy and difficult distractor tasks on the acquisition stage of memory. During easy distraction, brain activity associated with both episodic encoding processes and ones leading to priming in semantic memory will be observed. During difficult distraction only semantic processes occur. Thus, comparison of the two conditions identified brain activity associated with episodic encoding.

During retrieval, a task involving production of primed responses does not give any region of greater neural activation than an equivalent semantic memory task requiring only unprimed responses[5]. So experiment 2 (Table 1), which is concerned with episodic memory retrieval, also included a directly comparable task requiring only semantic memory retrieval (generation from a category such as furniture–table). Differences in activation should identify regions specifically associated with retrieval from episodic memory. Both experiments included control tasks designed to engage the non-semantic input and output demands of the experimental tasks (Table 1).

In the acquisition experiment, brain regions that were significantly more activated (showing greater regional cerebral blood flow) during encoding than during a passive listening task are shown in Figs 1*a* (easy distractor condition), *b* (difficult distractor condition) and Table 2. After difficult distraction, recall of the words was significantly reduced from 83 to 68%. Two brain areas were identified where there was marked activity during easy distraction (Fig. 1*a*), but no detectable activity during difficult distraction (Fig. 1*b*). From the logic of the experimental design, these two areas—left dorsolateral prefrontal and retrosplenial cortex—are specifically associated with episodic encoding processes. Figure 2*a* shows the regions in the retrieval experiment activated more by the recall task than a word repetition task; Fig. 2*b* shows the corresponding regions for the generation task. From the logic of the experiment, the two regions shown in Fig. 2*a* but not *b*—bilateral precuneus and right prefrontal cortex—are specifically associated with episodic memory retrieval (see also Table 2).

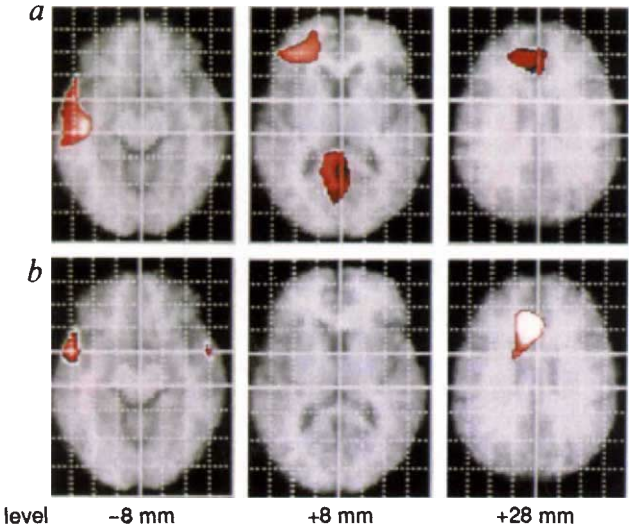


FIG. 2 Data presented as in Fig. 1. *a*, Regions significantly activated in the episodic memory retrieval condition (compared with word repetition). *b*, Regions significantly activated in the semantic memory condition. In both comparisons there is activation of the left anterior cingulate cortex, generally assumed to be associated with attention[27], and of the thalamus (bilaterally), long held to be implicated in memory functions[2]. In addition, relative deactivations (not shown) were observed for both comparisons in the superior and middle temporal lobes. Similar relative deactivations have been observed in a word-list recall study and in a verbal fluency task[10,12]. In such tasks attention is less directed towards auditory input processing than it is in the word repetition tasks with which they were compared. Activation of the precuneus bilaterally and right dorsolateral frontal cortex were observed only during retrieval from episodic memory.

The first important aspect of our results is that the same set of four regions were activated as in the only other study attempting to differentiate episodic memory from other memory processes, although in that study, acquisition and retrieval were conflated[10]. Particularly striking is the fact that not only were the tasks used in the earlier study very different—recall of supraspan and subspan word lists—but so also was the process contrasting with verbal episodic memory: auditory–verbal short-term memory rather than semantic memory. This convergence of evidence supports our claim that these four regions are involved in verbal episodic memory. Our observation that left frontal activity is associated with acquisition and right frontal activity with retrieval is also confirmed in other PET studies (E. Tulving *et al.*, personal communication).

The second important aspect is that the use of functional imaging has split these four regions into two involved with encoding and two with retrieval processes. The finding that the left dorsolateral prefrontal region is associated with encoding clarifies this important but poorly understood process[1]. This region is held to play a major role in the executive component of working memory[7,11,12] and is involved in the organization of supervisory thought processes[13,14]. The major reciprocal connections between the dorsolateral prefrontal region and the hippocampus involve the retrosplenial region[15], the other area activated in this study. Lesions to this region can produce amnesia[16,17]. These observations fit with the idea that encoding involves left frontal control of hippocampal function. For retrieval, the involvement of the right prefrontal area confirms the results of an earlier PET study of memory retrieval[5]. Furthermore, lesions producing the confabulatory disorders, observable after anterior communicating artery aneurgisms[18] and frontal head injury[19], involve similar areas of prefrontal cortex and occur more frequently after right- rather than left-sided damage[20]. Memory monitoring and verification are likely to be the critical processes impaired[14]. The involvement of the right frontal lobe is of special interest in that our task and that used in the previous PET study of memory retrieval[5] tested only verbal memory. Less clear is the function of the other area involved, the precuneus. Little is known about this structure from lesion studies. One possibility is that it is concerned with the retrieval of visual images[10,21]. This would be compatible with the known posterior cortical localization of visual imagery[22].

In common with nearly all relevant functional imaging studies[5,10], our study has failed to show selective activation of the medial brain structures (apart from the thalamus), damage to which causes amnesia[2]. Of these structures, the mamillary bodies may be too small to resolve. But the lack of selective activation of the hippocampus must be theoretically important[23]. Perhaps memory encoding in the hippocampus involves only very sparse neuronal activation[24]. Nevertheless, our study reveals a network of prefrontal and posterior cortical structures[6,7], with distinct roles in the different components of episodic memory. □

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Role of guanylyl cyclase and cGMP-dependent protein kinase in long-term potentiation

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SEVERAL lines of evidence suggest that cyclic GMP might be involved in long-term potentiation (LTP) in the hippocampus[1–6]. Arachidonic acid, nitric oxide and carbon monoxide, three molecules that have been proposed to act as retrograde messengers in LTP[7–9], all activate soluble guanylyl cyclase[1,10,11]. We report here that an inhibitor of guanylyl cyclase blocks the induction of LTP in the CA1 region of hippocampal slices. Conversely, cGMP analogues produce long-lasting enhancement of the excitatory postsynaptic potential if they are applied at the same time as weak tetanic stimulation of the presynaptic fibres. The enhancement is spatially restricted, is not blocked by valeric acid (APV), nifedipine, or picrotoxin, and partially occludes LTP. This synaptic enhancement may be mediated by the cGMP-dependent protein kinase (PKG). Inhibitors of PKG block the induction of LTP, and activators of PKG produce activity-dependent long-lasting enhancement. These results suggest that guanylyl cyclase and PKG contribute to LTP, possibly as activity-dependent presynaptic effectors of retrograde messengers.

An inhibitor of soluble guanylyl cyclase LY83583 (50% inhibitory concentration $IC_{50} = 2\,\mu M$)[12], completely blocked the induction of LTP at a concentration of 5 μM (Fig. 1*a*), and occasionally blocked it at a concentration of 0.5 μM (two of six experiments). The effects of the inhibitor are relatively specific. LY83583 (5 μM) did not affect either the maintenance of LTP (Fig. 1*a*), the baseline excitatory postsynaptic potential (e.p.s.p.; 96.1 ± 6.6%, $n = 9$), or the NMDA (*N*-methyl-D-aspartate) component of e.p.s.p. measured in the presence of 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX), which blocks the non-NMDA component (102 ± 14%, $n = 3$).

Conversely, application of the membrane-permeable analogue 8-Br-cGMP (100 μM) at the same time as weak tetanic stimulation (50 Hz, 0.5 s) ('paired' training) produced an immediate potentiation of the e.p.s.p. that lasted for at least 1 h (Fig. 1*c*). 8-Br-cGMP paired with weak stimulation produced significant enhancement even in the presence of 5 μM LY83583 (160.2 ± 5.9%, $n = 5$, $t[4] = 10.3$, $P < 0.01$, comparing the mean