

Amygdala Is Critical for Stress-Induced Modulation of Hippocampal Long-Term Potentiation and Learning

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Stress is a biologically significant factor shown to influence synaptic plasticity and memory functioning in the hippocampus. This study examined the role of the amygdala, a brain structure implicated in coordinating stress behaviors and modulating memory consolidation, in mediating stress effects on hippocampal long-term potentiation (LTP) and memory in rats. Electrolytic lesions of the amygdala effectively blocked the adverse physiological and behavioral effects of restraint and tailshock stress, without impeding the increase in corticosterone secretion to stress. Physiologically, hippocampal slices from stressed animals exhibited impaired LTP relative to slices from unstressed control animals, whereas hippocampal slices from stressed animals

with amygdalar lesions exhibited normal LTP. Behaviorally, stressed animals were impaired in retention of a hippocampal-dependent hidden platform version of the Morris water maze task, and this impairment was blocked by amygdalar lesions. In a fixed location–visible platform water maze task that can be acquired by independent hippocampal and nonhippocampal memory systems, stress enhanced the use of nonhippocampal-based memory to acquire the task. These results indicate that an intact amygdala is necessary for the expression of the modulatory effects of stress on hippocampal LTP and memory.

Key words: hippocampus; learning; fear; emotion; glucocorticoids; corticosterone; synaptic plasticity

It is well documented that adverse effects on cognitive functioning generally accompany stress (Maier and Seligman, 1976). Although the acute response to stress (e.g., heightened cognition) is an adaptive mechanism, excessive stress, in particular uncontrollable stress, can have severe repercussions ranging from impairments in learning and memory to enhanced susceptibility to neuronal cell death (for review, see McEwen and Sapolsky, 1995; Kim and Yoon, 1998).

The hippocampus, as part of a system necessary for the formation of stable memory (Scoville and Milner, 1957; Eichenbaum et al., 1992; Squire and Zola, 1996), is enriched with receptors for corticosteroids (the principal glucocorticoid secreted by the adrenal cortex in response to stress; cortisol in humans, corticosterone in rats) and participates in terminating the stress response via the glucocorticoid-mediated negative feedback of the hypothalamus–pituitary–adrenal axis (McEwen and Sapolsky, 1995). In the rat hippocampus, corticosterone has been shown to regulate metabolic, physiologic, and genomic functions of neurons (Sapolsky, 1992). As a result, certain hippocampal functions appear to be susceptible to stress, possibly linking the effects of glucocorticoids to cognitive functions such as learning and memory. For example, stress and corticosterone have been shown to impair hippocampal-dependent forms of verbal memory in humans (Bremner et al., 1993; Newcomer et al., 1999) and spatial memory in rats (Diamond et al., 1992; Luine et al., 1994; Bodnoff et al.,

1995; de Quervain et al., 1998). Consistent with these behavioral data, both *in vitro* and *in vivo* electrophysiological studies indicate that stress impairs hippocampal LTP (Foy et al., 1987; Shors et al., 1989; Diamond et al., 1992; Shors and Dryver, 1994; Kim et al., 1996; Xu et al., 1997), a putative cellular mnemonic mechanism (Morris et al., 1990; Bliss and Collingridge, 1993) (but see Shors and Matzel, 1997). If the notion that changes in synaptic efficacy are essential for learning and memory [e.g., Hebb's postulate; Hebb (1949)] is correct, then it is possible that the LTP impairment associated with stress might be one neural basis for stress-induced alterations in learning.

Considerable evidence indicates that the amygdala is critically involved in mediating stress-related effects on behavior and modulating hippocampal function. For example, amygdalar lesions and/or pharmacological manipulations have been shown to (1) prevent stress-induced gastric erosion (Henke, 1981, 1990) and analgesia (Helmstetter, 1992), (2) block memory modulatory effects of intrahippocampally administered drugs (Roosendaal et al., 1996, 1998; Packard and Chen, 1999), and (3) impair *in vivo* dentate gyrus LTP in the hippocampus (Ikegaya et al., 1994, 1995, 1996). In addition, the amygdala has been implicated in emotional learning (Kim et al., 1993; LeDoux, 1994; Maren and Fanselow, 1996) and attention (Gallagher and Schoenbaum, 1999; Holland et al., 2000). Anatomically, the amygdala projects to several hippocampal regions (including the CA1 area) (Krettek and Price, 1977; Aggleton, 1986), providing various routes by which it may potentially influence hippocampal function. Therefore, the present series of experiments examined the possibility that the amygdala is involved in mediating stress effects on hippocampal LTP and hippocampal-dependent learning, using a hidden platform version of the Morris water maze task. In view of evidence that memory is organized in multiple brain systems (Packard et al., 1989; Squire and Zola, 1996; Thompson and Kim, 1996), we also examined whether stress might influence learning in a

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task in which both hippocampal-dependent and hippocampal-independent memory systems appear to be engaged (McDonald and White, 1994). Specifically, we hypothesized that a selective impairing effect of stress on hippocampal memory processes would enhance the use of hippocampal-independent memory in acquiring this task.

MATERIALS AND METHODS

Subjects. Experimentally naïve male Charles River Long–Evans rats (270–300 gm) were individually housed in a climate-controlled vivarium on a 12 hr light/dark cycle (lights on at 7:00 A.M.) with *ad libitum* access to food and water. The animals were handled daily for 7 d before surgery. All experiments were conducted during the light phase of the cycle.

Surgery. Under ketamine HCl (30 mg/kg) and xylazine (2.5 mg/kg) anesthesia, subjects were mounted in a stereotaxic instrument (Stoelting, Wood Dale, IL), and bilateral amygdalar lesions were made by passing constant current (1.5 mA, 15 sec; Ugo Basile, Comerio, Italy) through a stainless steel insect pin (#00) that was insulated with epoxy, except for ~0.5 mm at the tip (coordinates: from bregma, –2.3 mm posterior, ±4 and ±5 mm lateral, and –8.4 and –8.8 mm ventral from the skull) (cf. Kim et al., 1993). For operated sham controls, the electrode was lowered to the amygdala without passing current. After surgery, all animals were given between 2 and 5 weeks to recover and acclimate to daily handling.

Stress paradigm. Half of the animals from sham and lesion groups were restrained in a Plexiglas tube and exposed to 60 tailshocks (1 mA intensity, 1 sec duration, 30–90 sec variable intershock interval), whereas the remaining animals were left undisturbed (four groups: sham-control, sham-stress, lesion-control, lesion-stress). This stress procedure, adapted from the “learned helplessness” paradigm (in which animals undergo an aversive experience under conditions in which they cannot perform any adaptive response) (Seligman and Maier, 1967; Maier and Seligman, 1976), has been demonstrated to be effective in altering subsequent synaptic plasticity in the hippocampus (Foy et al., 1987; Shors et al., 1989; Kim et al., 1996).

In vitro electrophysiology procedure. Promptly after stress, animals were decapitated under halothane anesthesia, and hippocampal slices were prepared in a standard manner (cf. Teyler, 1980). In brief, transverse hippocampal slices (400 μ m) were maintained in an interface recording chamber (Fine Science Tools, Foster City, CA) and continuously perfused (~2 ml/min) with 95% O₂ and 5% CO₂ saturated artificial CSF [(in mM) 124 NaCl, 3 KCl, 1.25 NaH₂PO₄, 1 MgSO₄, 26 NaHCO₃, 3 CaCl₂, and 10 glucose] at 32°C. After at least 1 hr of incubation, a concentric bipolar electrode (inner contact diameter, 25 μ m) delivering 100 μ sec pulses stimulated the Schaffer collateral–commissural fibers. A glass electrode filled with 2 M NaCl (1.5–2.5 M Ω) was placed in the stratum radiatum in CA1 under a microscope to record field EPSPs (f-EPSPs). The test stimulus intensity was adjusted to produce a response that was 50% of the maximum evoked responses. Baseline synaptic transmission was monitored for 20 min (every 20 sec) before delivering a tetanus [five trains of 100 Hz, each lasting 200 msec at an intertrain interval (ITI) of 10 sec]. The f-EPSPs (amplified in the band of 0.1–5000 Hz) were monitored up to 1 hr after the tetanus. During the tetanus, f-EPSPs that were evoked by the first pulse in each of the five trains were recorded to assess the development of potentiation. Data were collected and analyzed on-line using a computer program written in AxoBasic/QuickBasic (Axon Instruments, Foster City, CA). The initial (negative) slope of f-EPSPs was used in statistical analyses (cf. Kim et al., 1996). Only those slices that exhibited a stable baseline for 20 min were included in the analysis. The change in f-EPSPs after tetanus was averaged across slices for each rat (usually two hippocampal slices per rat). The magnitude of LTP was measured between 40 and 60 min after the tetanus, and statistical comparisons were made to the 20 min baseline measure.

Corticosterone radioimmunoassay. During the hippocampal slice preparation, trunk blood was collected for corticosterone radioimmunoassay. Blood serum was separated by centrifugation (5000 rpm, 20 min) and stored at –80°C until the time of assay. Serum corticosterone was measured using the radioimmunoassay kit of ICN Biomedicals (Carson, CA) with ¹²⁵I-corticosterone as a tracer.

Hidden platform water maze task. The training and testing procedures were adapted from those previously described and have been shown to be hippocampal-based (Packard and McGaugh, 1994; Packard and Teather, 1998). After stress or not (in the manner described above), experimentally naïve amygdalar lesion and sham animals were placed back in their home cages for 30–60 min before undergoing eight massed training trials

(1 min ITI) to find a fixed submerged platform and escape from a circular water maze (diameter, 2.0 m; height, 0.7 m; water temperature, 23°C). The starting point was randomly distributed across the four quadrants (two starting points per quadrant; the animal always faced the wall when placed in the water). If escape did not occur within 60 sec, the animal was manually guided to the platform. On finding the platform, the animal remained on the platform for 30 sec and then was placed in a holding cage for another 30 sec before the next trial. After the last trial, the animals were returned to their home cages. The next day, a retention test (a 60 sec probe trial) was given in which the platform was removed from the pool. Animals' movements and the time taken to reach the position at which the platform had been located in training were monitored automatically using a computerized Poly-Track Video Tracking System (San Diego Instruments, San Diego, CA).

Two days after the spatial memory test, all animals were tested for fear conditioning to validate the functional effectiveness of the amygdalar lesions. After 3 min of baseline in a modular operant test cage (Coulbourn Instruments, Allentown, PA), animals were presented with three un signaled footshocks (1 mA, 1 sec, 1 min intershock interval) through the floor grid, which was wired to a Coulbourn precision-regulated animal shocker. Fear conditioning was monitored during the three 1 min intershock intervals by measuring freezing behavior using a 24-cell infrared activity monitor that detects movement of an emitted infrared (1300 nm) body heat image from the animal in the horizontal and vertical planes (cf. Lee and Kim, 1998). Immediate postshock freezing has been shown to accurately assay fear conditioning (Kim et al., 1992, 1993).

At the completion of behavioral testing, the subjects were overdosed with ketamine HCl and xylazine and perfused intracardially with 0.9% saline, followed by 10% buffered formalin. The brains were removed and stored in 10% formalin for at least 2 weeks before slicing. Transverse sections (60 μ m) were taken through the extent of the lesion, mounted on gelatinized slides, and stained with cresyl violet and Prussian blue dyes.

Fixed location–visible platform water maze task. The training and testing procedures were adapted from McDonald and White (1994), who demonstrated that this task is acquired by independent hippocampal-based and dorsal striatal-based memory systems. Naïve animals (no surgery) were exposed to stress or not and then placed back in their home cages for 30–60 min before undergoing eight massed trials (1 min ITI) to find a fixed submerged platform coupled with a visually salient pole (a black and white striped plastic strip; 15.2 \times 1.2 cm). As with the hidden platform task, the starting point was randomly distributed across the four quadrants. On finding the platform, the animal remained on the platform for 30 sec to explore its surroundings and then was placed in a holding cage for another 30 sec before the next trial. After the last trial, animals were placed back in their home cages. The next day, a retention test was given in which the platform (coupled with the pole) was moved to a novel location. Animals were placed into the pool facing the wall equidistant from the previous and new platform locations, and their movements were tracked.

RESULTS

Figure 1 shows a photomicrograph of a transverse brain section stained with cresyl violet and Prussian blue from a typical rat with bilateral electrolytic lesions in the amygdala. Amygdaloid damage typically involved the majority of central and basolateral–lateral nuclei and small portions of the amygdala–striatal transition area.

As shown in Figure 2*A*, hippocampal slices from sham-stress animals exhibited impaired LTP (normalized f-EPSP slopes measured 40–60 min after the tetanus: 107.2 \pm 6.3%), whereas LTP was robust in slices from sham-control (149.2 \pm 3.0%), lesion-control (140.9 \pm 4.7%), and lesion-stress (139.4 \pm 6.5%) animals (two-way ANOVA; lesion \times stress interaction: $F_{(1,27)} = 11.5, p < 0.01$; planned comparisons: $F_{(3,27)} = 8.5$, all p values < 0.01 Newman–Keuls). There was a significant main effect of stress on LTP ($F_{(1,27)} = 10.4, p < 0.01$), but no reliable main effect of lesion on LTP ($F_{(1,27)} = 3.6, p > 0.05$), indicating that the amygdalar lesions did not reduce the magnitude of LTP per se but did effectively block stress-induced impairments in LTP.

Examination of blood corticosterone levels (Fig. 2*B*) revealed significantly higher levels in animals exposed to stress than in those not exposed to stress, irrespective of the lesion, (two-way

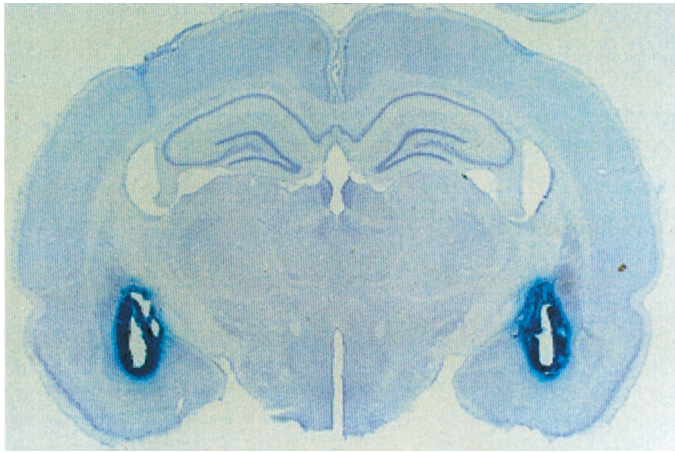


Figure 1. Photomicrograph showing a transverse brain section stained with cresyl violet and Prussian blue from a rat with amygdalar lesions.

ANOVA; main effect of stress: $F_{(1,27)} = 78.9, p < 0.01$; main effect of lesion: $F_{(1,27)} = 2.3, p > 0.05$; lesion \times stress interaction: $F_{(1,27)} = 1.3, p > 0.05$). Although there appears to be a trend of lesion-stress animals ($49.7 \pm 7.8 \mu\text{g/dl}$) showing a lesser amount of stress-induced corticosterone elevation than sham-stress animals ($65.0 \pm 7.6 \mu\text{g/dl}$), this difference was not statistically reliable, ($p > 0.05$, Newman–Keuls). This indicates that amygdalar lesions do not affect stress-induced elevations in corticosterone levels.

In a hippocampal-dependent hidden platform version of the water maze task, all groups significantly decreased their latencies to find the hidden platform during the eight training trials (Fig. 3A). The rate of acquisition was comparable among the four groups (two-way ANOVA with trials as a repeated measure; main effect of lesion: $F_{(1,31)} < 1.0, p > 0.05$; main effect of surgery: $F_{(1,31)} = 2.7, p > 0.05$; lesion \times stress \times trials interaction: $F_{(7,245)} < 1.0, p > 0.05$). On the retention (probe) test a day later, however, the lesion animals required significantly shorter latencies to swim to the original location of the platform than the sham animals, irrespective of stress (two-way ANOVA; $F_{(1,34)} = 13.5, p < 0.01$). Although neither the main effect of stress nor lesion \times stress interaction was significant (two-way ANOVA; $F_{(1,34)} = 3.0, p > 0.05$, and $F_{(1,34)} = 1.4, p > 0.05$, respectively), a simple planned comparison analysis indicated that the sham-stress animals ($39.1 \pm 8.4 \text{ sec}$) exhibited significantly longer latencies to

swim to the original location of the platform, in comparison to the sham-control ($22.7 \pm 3.9 \text{ sec}$), the lesion-control ($9.8 \pm 2.5 \text{ sec}$), and the lesion-stress ($12.9 \pm 4.7 \text{ sec}$) animals (one-way ANOVA; $F_{(3,34)} = 6.1$, all p values < 0.05 Newman–Keuls). The latency differences cannot be attributed to possible motoric effects because there were no reliable group differences in swim speed ($p > 0.05$) (Fig. 3B). Also, the swim distance-dependent measure provided the same results as the latency-dependent measure (data not shown). Thus, these results suggest that amygdalar lesions may enhance retention of the hidden platform task and also effectively block the impairing effects of stress on this task.

Using the present training parameters in which rats are trained in a single rapid session (eight trials per 60 sec ITI), none of the groups subsequently demonstrated a reliable quadrant bias or difference in number of quadrant entries per annulus crossing on the probe trial given 24 hr later (data not shown). However, on the probe trial, the sham-stress animals had significantly longer latencies to reach the location of the platform and swam significantly longer distances to reach this location, compared with sham-control, lesion-control, and lesion-stress animals. Importantly, the significance in the latency measure was not attributable to a difference in swim speed between the four groups of animals. Although a reliable spatial bias was not observed in unstressed animals during the probe trial, it should be noted that previous drug infusion studies (Packard et al., 1994; Packard and Teather, 1998) have shown the current training and testing procedures (using the latency and distance measures) to be hippocampal-based.

In addition to histological verification (Fig. 1), the effectiveness of the lesions was confirmed by observations that animals with amygdalar lesions exhibited virtually no freezing after footshocks (main effect of lesion: $F_{(1,34)} = 171.8, p < 0.01$) (Fig. 3B). Previous experience with stress (3 d before fear conditioning) did not reliably affect fear conditioning in either lesion or sham groups (no lesion \times stress interaction: $F_{(1,34)} = 0.02, p > 0.05$).

In the fixed location–visible platform water maze task, an ANOVA with group as a between-subject factor and trials as a repeated measure revealed that there was no reliable difference between stress and control animals during the eight acquisition trials ($F_{(7,140)} = 1.5, p > 0.05$) (Fig. 4A). On the retention test (24 hr later), with the platform (marked with the same salient pole) moved to a new location, animals stressed before training exhibited a significantly shorter latency to swim to the platform than unstressed control animals ($F_{(1,19)} = 4.7, p < 0.05$) (Fig. 4A). The control animals exhibited longer latencies to escape because they

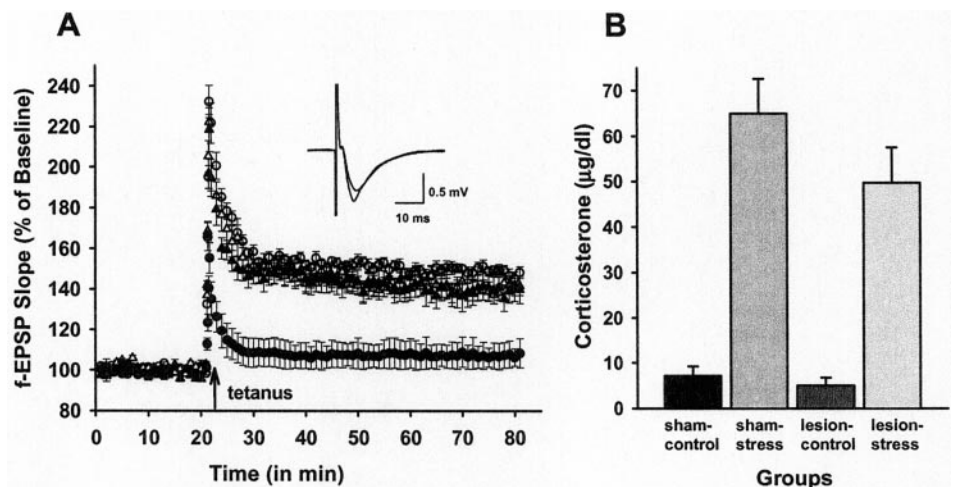


Figure 2. Effects of amygdalar lesions and stress on Schaffer collateral–commissural-CA1 LTP and corticosterone secretion. *A*, Synaptic strength in the CA1 area of the hippocampus from sham-control (open circles, $n = 6$), sham-stress (filled circles, $n = 6$), lesion-control (open triangles, $n = 8$), and lesion-stress (filled triangles, $n = 8$) animals is expressed as a percentage of the average pre-tetanus f-EPSP over time (in minutes). *Inset*, two representative f-EPSPs from lesion-stress group, taken 10 min before and 50–60 min after LTP are shown. *B*, Trunk blood corticosterone levels assayed (^{125}I radioimmunoassay) from the four groups used in the hippocampal slice experiment.

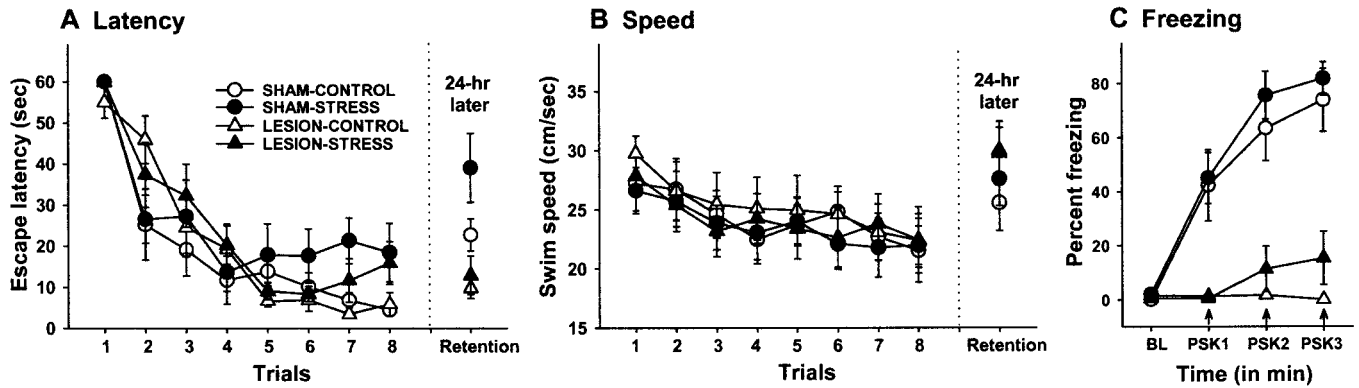


Figure 3. Effects of amygdalar lesions and stress on spatial memory and fear conditioning. *A*, Mean (\pm SE) latencies to find a submerged platform from sham-control (open circles, $n = 8$), sham-stress (filled circles, $n = 9$), lesion-control (open triangles, $n = 9$), and lesion-stress (filled triangles, $n = 9$) animals during acquisition and a single retention test. *B*, Mean (\pm SE) swim speed (centimeters per second) of four groups during acquisition and a single retention test. *C*, Mean (\pm SE) percentage postshock (PSK) freezing during the 1 min baseline (BL) and during the three 1 min intershock intervals.

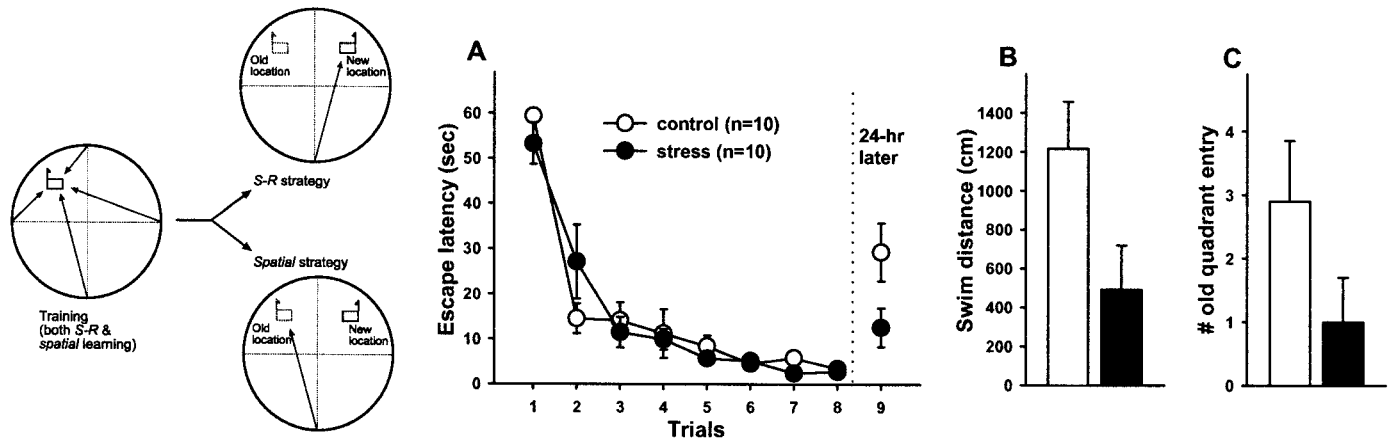


Figure 4. Left, Fixed location-visible platform water maze paradigm for assessing stress effects on the relative use of S-R and spatial memory. *A*, Mean (\pm SE) latency to find a submerged platform marked with a visually salient pole from control (open circles, $n = 10$) and stress (filled circles, $n = 10$) animals during the acquisition trials (1–8) and on a single test trial (9). *B*, Mean (\pm SE) distance to find a submerged platform marked with a visually salient pole on a single test trial. *C*, Mean number of old quadrant entry (where the platform was located during training).

(10 of 10) initially swam to the original platform location (preferentially using a spatial strategy) before swimming to the visible platform now located in a new quadrant. In contrast, 5 of 10 stress animals swam directly to the new platform location [preferentially using a stimulus–response (S-R) strategy], whereas the remaining 5 animals swam to the original platform location before the new platform location (preferentially using a spatial strategy). The swim distance to the new platform location and the number of old quadrant entry measures (Fig. 4*B,C*) also indicate that stress enhances the use of an S-R strategy in this task.

DISCUSSION

The present findings demonstrate that amygdalar lesions effectively block stress effects on hippocampal LTP and hippocampal-dependent memory and are consistent with previous reports that amygdalar lesions prevent other effects of stress, including gastric erosion (Henke, 1990) and analgesia (Helmstetter, 1992). Specifically, we found that hippocampal slices obtained from sham animals exposed to stress exhibited LTP impairments in the CA1 area, whereas slices from sham animals not exposed to stress demonstrated robust LTP, replicating earlier *in vitro* and *in vivo* findings of stress-induced impairment of LTP (Foy et al., 1987; Shors et al., 1989; Diamond and Rose, 1994; Kim et al., 1996; Xu

et al., 1997). In contrast, LTP was observed reliably in hippocampal slices prepared from amygdala-lesioned animals, regardless of whether or not they experienced stress. Similarly, we observed that amygdalar lesions also blocked stress-induced memory impairments when rats were tested in a hidden platform water maze task that has previously been shown to be hippocampus-based (Packard et al., 1994; Packard and Teather, 1998). Thus, our findings that the amygdala is critically involved in mediating stress effects on hippocampal LTP and hippocampal-dependent memory are consistent with the view that one function of the amygdala is to modulate memory processes in other brain structures, such as the hippocampus (Gallagher and Kapp, 1978; Ikegaya et al., 1994; Packard et al., 1994; Cahill and McGaugh, 1998; Packard and Teather, 1998; Roozendaal et al., 1998; Packard and Chen, 1999; McGaugh, 2000).

In the present study, sham lesion animals exposed to 1 hr of uncontrollable stress (60 tailshocks and restraint) before undergoing water maze training (pretraining stress effects) exhibited impairments in spatial memory when tested 24 hr later. In another study (de Quervain et al., 1998), a relatively milder three footshock stress (lasting <1 min) that was presented before a retention test (pretesting stress effects) impaired performance in

a water maze spatial task in a time-dependent manner (i.e., retention was impaired 30 min poststress but not 2 min or 4 hr poststress) that corresponds to the corticosterone levels at the time of testing. It appears then that pretraining exposures to a relatively intense and longer-lasting stress (used in the present study) can affect spatial memory in a manner that does not directly correspond to the corticosterone levels at the time of testing (24 hr later).

Because there is no evidence that three footshock stress influences hippocampal plasticity (i.e., LTP), it would be important to investigate whether or not these two different magnitudes of stress produce similar pretraining and pretesting effects on hippocampal-dependent memory.

Interestingly, using the present training–testing procedures, lesioning the amygdala per se seems to enhance the performance in the hidden platform water maze task. This finding differs from a previous study (Sutherland and McDonald, 1990) that found neither enhancing nor impairing effects of amygdalar lesions on a spatial version of the water maze task when animals were trained across several days. It is conceivable that high levels of stress hormones (such as epinephrine and glucocorticoids) are released during the eight massed water maze training trials, which might normally produce memory impairing effects in the amygdala-intact animals. Thus, this finding is consistent with the accumulating evidence indicating that amygdala function is necessary for intrahippocampally administered drugs to modulate (enhance or impair) consolidation of hippocampal-dependent (e.g., spatial) memory and for mediating memory modulatory effects of stress hormones (Cahill and McGaugh, 1991; Packard et al., 1994; Roozendaal and McGaugh, 1996, 1997; Roozendaal et al., 1998; Packard and Teather, 1998; Packard and Chen, 1999; McGaugh, 2000).

It is also significant that amygdalar lesions did not affect Schaffer collateral–commissural-CA1 LTP in hippocampal slices from unstressed animals. Recent studies suggest that the amygdala influences LTP in the hippocampus. For instance, electrolytic lesions to the basolateral (but not central) nuclei of the amygdala have been shown to significantly attenuate perforant path–dentate gyrus LTP *in vivo* (Ikegaya et al., 1994), whereas high-frequency stimulation of the amygdala augmented LTP (Ikegaya et al., 1996). It now appears that stimulation of the amygdala induces a time-dependent biphasic effect on hippocampal LTP (an immediate excitatory effect and a longer-lasting inhibitory effect) (Akirav and Richter-Levin, 1999). Additionally, intra-amygdala infusions of NMDA receptor antagonists have been found to impair dentate gyrus LTP (without affecting the baseline synaptic response), suggesting that NMDA receptors in the amygdala might be involved in influencing LTP (Ikegaya et al., 1995). In the present study, however, although amygdalar lesions (which included both central and basolateral nuclei) blocked stress effects on CA1 LTP *in vitro*, the lesions did not affect LTP in unstressed animals. Thus, it is possible that the amygdala may differentially influence synaptic plasticity in different regions of the hippocampus.

Although stress impaired retention of hippocampal-dependent memory in a hidden platform water maze task, the same stress enhanced the relative use of hippocampal-independent S-R memory in a fixed location–visible platform water maze task in which both hippocampal-dependent and caudate-dependent memory systems are engaged (McDonald and White, 1994). The effects of stress on behavior in this task are similar to those of fornix lesions, which also result in enhanced use of S-R behavior relative

to normal animals (McDonald and White, 1994). Thus, both stress (presumably via impairing hippocampal LTP) and fornix lesions (via disrupting hippocampal afferent-efferent pathways) impair the use of spatial information and facilitate the use of S-R information in the acquisition of an escape response to a visible platform in a fixed location. Similarly, stress (Shors et al., 1992; Shors and Mathew, 1998) and hippocampal lesions (Schmaltz and Theios, 1972; Port et al., 1985) have been shown to facilitate the acquisition of hippocampal-independent (but cerebellar-dependent) delay eyeblink conditioning (Kim et al., 1995; Kim and Thompson, 1997). It has also been reported that infusions of NMDA receptor antagonists into the amygdala before stress effectively block stress-induced facilitation of eyeblink conditioning (Shors and Mathew, 1998). Thus, it would be important to test whether NMDA receptor antagonists in the amygdala would also block stress-induced enhancement of hippocampal-independent S-R memory as well as stress-induced impairment in hippocampal LTP and spatial memory. At any rate, our findings are consistent with the general notion that amygdala activation can influence both hippocampal-dependent and hippocampal-independent memory (Packard et al., 1994; McGaugh, 2000).

It is generally viewed that there are multiple memory systems that are subserved by different brain substrates (Packard et al., 1989, 1994; Packard and McGaugh, 1992; Squire and Zola, 1996; Thompson and Kim, 1996). Under normal conditions, however, competition for control of learned behavior may arise among these systems. For example, although the hippocampus is not essential for delay eyeblink conditioning (Schmaltz and Theios, 1972; Kim et al., 1995), hippocampal lesions can facilitate the acquisition of delay eyeblink conditioning (Port et al., 1985), pretraining LTP saturation in the hippocampus accelerates the rate of delay eyeblink conditioning (Berger, 1984), and PKC γ mutant mice (deficient in the γ isoform of protein kinase C) with a moderate impairment in hippocampal LTP (Abeliovich et al., 1993) exhibit facilitated acquisition of delay eyeblink conditioning (Chen et al., 1995). In addition, lesions of the hippocampal system facilitate the acquisition of caudate-dependent S-R learning in a win-stay radial maze task (Packard et al., 1989; McDonald and White, 1993). Together, these results indicate that during hippocampal-independent learning (e.g., delay eyeblink conditioning, S-R learning), the hippocampus may be engaged in processing information (e.g., context) (Good and Honey, 1991; Kim and Fanselow, 1992; Phillips and LeDoux, 1992) that might interfere with the formation or expression of hippocampal-independent memory. Thus, stress-induced alterations in synaptic plasticity that selectively affect hippocampal memory processes may inhibit the competitive interference between hippocampal-dependent and hippocampal-independent memory systems and thereby enhance performance in nonhippocampal learning tasks.

With regard to stress effects on hippocampal LTP, it has been reported previously that there is a biphasic relationship between level of corticosterone and magnitude of LTP (Diamond et al., 1992), with both low (via adrenalectomy) and high (via exogenous administration) levels of corticosterone impairing LTP. In addition, corticosterone has been shown to affect the intrinsic properties of hippocampal neurons (e.g., prolonging the afterhyperpolarization) (Joels and De Kloet, 1989; Kerr et al., 1989) that would reduce cell excitability. Behaviorally, rats that were administered corticosterone at doses comparable with those observed during natural stress were found to be impaired in spatial learning (Bodnoff et al., 1995). Given these findings, it is surprising that amygdalar lesions effectively blocked stress effects on hippocam-

pal LTP and spatial memory without significantly affecting the increase in corticosterone secretion in response to stress. Our results suggest that this increase in corticosterone levels is not a sufficient condition to mediate stress effects on hippocampal plasticity and learning. This view is also supported by findings that LTP is reduced further in adrenalectomized rats after stress and is not restored by exogenous administration of corticosterone (Shors et al., 1990), and that in normal animals administered with dexamethasone (a synthetic glucocorticoid that blocks the HPA axis activity), stress-induced impairments in LTP nonetheless occurred (Foy et al., 1990). Collectively, these data indicate that multiple factors (in addition to glucocorticoids) mediate stress effects on hippocampal functioning.

In conclusion, the current findings suggest that alterations in hippocampal plasticity subsequent to stress might be caused by excessive modulatory effects of the amygdala during the stress experience. If amygdalar modulation of hippocampal physiology occurs during stress, then this effect must have a long duration because it was observed in hippocampus *in vivo*. It is now of interest to characterize the neuroanatomical–neurochemical projections from the amygdala to the hippocampus to further elucidate the modulating mechanisms of stress on neural plasticity and memory processes.

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