Individual differences in emergence neophobia predict magnitude of perforant-path long-term potentiation (LTP) and plasma corticosterone levels in rats

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Emergence neophobia was assessed in an emergence apparatus that provided a choice between novel and familiar alternatives. Two weeks after emergence testing, the threshold to induce perforant-path long-term potentiation (LTP) and the magnitude of perforant-path LTP in the dentate gyrus were assessed under pentobarbital anesthesia. Two measures of emergence behavior, the total duration of time spent in the alley during the 1-h test and the emergence duration per entry into the novel compartment, were significantly correlated with LTP of the extracellular population excitatory postsynaptic potential (EPSP), but not with the population spike. Neophobic animals that spent relatively little time in the novel alley during the 1-h test had a lower threshold to induce LTP and exhibited greater asymptotic EPSP LTP than did neophilic animals that readily entered and explored the novel alley. In a second experiment, plasma corticosterone levels in animals tested in the emergence task were also correlated with emergence duration and were generally lower in neophobic animals. Together, these data suggest that neotic behavior and LTP share a common mechanism, possibly one mediated by an interaction of glucocorticoid hormones and habituation.

Long-term potentiation (LTP) is an enduring form of synaptic enhancement induced at excitatory synaptic contacts in the mammalian brain by brief episodes of rhythmic electrical stimulation (Bliss & Lomo, 1973; Larson & Lynch, 1986). In the hippocampus, LTP exhibits many properties characteristic of memory, including rapid induction, temporal persistence, and associativity (Bliss & Lynch, 1988; Morris, Davis, & Butcher, 1990; Teyler & DiScenna, 1984). Several investigators have shown that 1) manipulations that disrupt hippocampal LTP can influence learning in a variety of tasks (Berger, 1984; Kim, DeCola, Landeira-Fernandez, & Fanselow, 1991; McNaughton, Barnes, Rao, Baldwin, & Rasmussen, 1986; Mondadori, Weiskrantz, Buerki, Petschke, & Fagg, 1989; Morris, Anderson, Lynch, & Baudry, 1986; Robinson, Crooks, Shinkman, & Gallagher, 1989; Shapiro & Caramanos, 1990; Staubli, Thibault, DiLorenzo, & Lynch, 1989), 2) LTP-like changes occur in the hippocampus during learning (Skelton, Scarth, Wilkie, Miller, & Phillips, 1987; Weisz, Clark, & Thompson, 1984), and

3) LTP-inducing stimulation can serve as an effective conditional stimulus (LaRoche, Doyere, & Bloch, 1989). Collectively, these data have fostered the view that hippocampal LTP plays an important role in learning.

A recent report indicates that novel stimuli without explicit associational contingencies can modulate the induction of LTP (Diamond, Bennett, Stevens, Wilson, & Rose, 1990). Simply exposing an animal to a novel environment considerably reduced the incidence of primed-burst potentiation in hippocampal area CA1, whereas habituation to the novel environment typically restored this form of LTP (Diamond et al., 1990). This novelty-related suppression of LTP has been linked to elevated levels of serum corticosterone that accompany forced exposure to novelty (Bennett, Diamond, Fleshner, & Rose, 1991) and is consistent with several reports of the inhibitory effects of exogenous stressors on LTP induction (Foy, Stanton, Levine, & Thompson, 1987; Shors, Seib, Levine, & Thompson, 1989; Shors & Thompson, 1992).

The sensitivity of hippocampal LTP to novel environmental stimuli suggests that it may be an important component of a hippocampal system involved in orchestrating behavioral responses to the relative novelty/familiarity of environmental stimuli. These responses, collectively referred to as "neotic behavior" (Corey, 1978), include orientation, exploratory approach, and neophobic avoidance. Many studies have shown that neotic responses are mediated by both experiential and dispositional factors. It is well established that habituation, which can be viewed as a process that converts novel stimuli into familiar stim-

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uli, plays a crucial role in exploratory behavior (Berlyne, 1960; Bindra, 1959; Corey, 1978; Dember & Earl, 1957; Welker, 1961). Similarly, exploratory tasks are notoriously sensitive to stress and arousal levels (Aitken, 1972; Bronson, 1968; Halliday, 1967; Montgomery, 1955; Russell, 1973; Williams, 1972). Moreover, habituation and arousal have been shown to interact in mediating neotic choice behaviors in a variety of behavioral tasks providing clear distinctions between novel and familiar alternatives (Mitchell, Fairbanks, & Laycock, 1977; Mitchell, Kirschbaum, & Perry, 1975; Mitchell, Koleszar, & Scopatz, 1984; Mitchell, Osborne, & O'Boyle, 1985; Sheldon, 1968).

That LTP participates in neotic behavior is congruent with a substantial portion of the literature on the functional role of the hippocampus. A number of studies have investigated the effects of hippocampal damage on the exploration of novel environments, distractibility in familiar environments, and related behaviors such as T-maze alternation (see Gray & McNaughton, 1983; Isaacson, 1974, and O'Keefe & Nadel, 1978, for reviews). Though consensus is elusive, several authors have concluded that the hippocampus plays an important functional role in neurophysiological processes that are thought to mediate exploration and habituation (Douglas, 1967; Gaffan, 1972; Kimble, 1968; Maren, Hwang, & Mitchell, 1992; Schmajuk, 1984). This conclusion is supported by several recent reports showing that (1) hippocampal structural traits correlate with habituation to novel environments (Crusio & Schwegler, 1987; Lipp, Schwegler, Heimrich, Cerbone, & Sadile, 1987; Patacchioli, Tagliabata, Angelucci, Cerbone, & Sadile, 1989), (2) preventing LTP induction disrupts habituation in a spatial novelty task (Carnevale, Vitullo, & Sadile, 1990), and (3) exploration and environmental enrichment are associated with LTP-like increases in hippocampal extracellular field potentials (Green, McNaughton, & Barnes, 1990; Sharp, McNaughton, & Barnes, 1985, 1989).

All of the above evidence suggests that the neotic information processing that occurs during exploration of novel environments may be modulated by hippocampal synaptic plasticity. In this report, we present two experiments that address this possibility. In Experiment 1, we show that neotic behavior in an emergence task predicts both the threshold to induce and the magnitude of perforant-path granule-cell LTP in anesthetized rats. In Experiment 2, we present complementary data indicating that emergence behavior is highly correlated with basal plasma corticosterone levels. Together, these data suggest that neotic behavior and LTP share a common mechanism, possibly one mediated by an interaction of glucocorticoid hormones and habituation.

METHOD

Subjects

Twenty male Long-Evans rats (Simonsen) approximately 80 days old (286–353 g) were individually housed in an air-conditioned room on a 12:12-h light:dark cycle (lights on at 0600 h). Each animal

occupied a standard stainless steel isolation cage (24.0 × 17.4 × 18.5 cm) suspended over a stainless steel tray covered with a layer of woodchips. Food (Purina Lab Chow) was continuously available from stainless steel hoppers attached to the back of each cage. Tap water was continuously available from glass bottles with stainless steel spouts attached to the front of each cage. The animals were transported to and from the emergence apparatus, which was located in an adjacent room, in their home cages.

Emergence Apparatus and Procedure

The emergence apparatus consisted of a set of four parallel wooden alleys (90.5 × 20.0 × 28.5 cm) painted gray and covered with hardware cloth (5.1 × 2.5 cm mesh). Four identical nest boxes (24.0 × 20.0 × 28.5 cm) with hinged hardware cloth tops (0.5-cm mesh) were separated from the alleys by guillotine doors forming the front wall of each box. The nest boxes and doors were painted black. Each box was ventilated through a 4.0-cm-diam grated hole centered 9.0 cm below the top of each back wall. The ventilation holes accessed a common manifold connected by flex tubing to a squirrel cage exhaust fan mounted outside the enclosure. A 15.0-cm-wide black wooden shelf mounted 4.5 cm above the nest boxes partially shaded the back portion of each box and supported a digital clock and animal identification cards. Food (Purina Lab Chow) and woodchip bedding were supplied on the floor of each nest box. Water was available from an externally mounted glass bottle with a stainless steel spout that projected through the back wall of the nest box 6.0 cm above the floor. The apparatus was positioned on the floor of a small enclosure (135.0 × 104.0 × 155.0 cm), with a rectangular opening (90.5 × 80.5 cm) in the center of the top and a sliding door along one side. Indirect lighting was provided by four fluorescent tubes (20 W) mounted vertically on the walls of the enclosure, two behind and two in front of the apparatus. A monitor and video cassette recorder were positioned on a cart outside the enclosure behind the apparatus, and a video camera, mounted on scaffolding 260.0 cm above the floor, permitted a clear view of the entire apparatus.

The animals were individually housed for 10 days before the beginning of the experiment, when they were run in the emergence task in five consecutive sets of 4 rats each. Twenty-four hours before each test (1800 h) the appropriate animals were placed in the nest boxes adjacent to the alleys. Food and water were freely available in the nest boxes, and the lighting cycle was the same as that in the vivarium where the animals were housed. The test was conducted at the beginning of the dark cycle the following day. At the beginning of each test, the enclosure lights were reset to remain on during the test, the camera was turned on, and the guillotine doors were removed. The animals were permitted to enter and explore the novel alleys for 1 h. Following the test, the animals were returned to their home cages. After each set of animals had been tested, the soiled litter and uneaten food were discarded and the entire apparatus thoroughly cleaned and washed with a mild detergent solution. The water bottles were cleaned and refilled, and each nest box was resupplied with fresh food and woodchip bedding. Subsequent sets of animals were housed in the nest boxes on the following day, and the procedure was repeated until all five sets had been run. The videotapes were scored for the latency to enter the alley (emergence latency, EL), the number of alley entries (emergence entries, ENT), and the duration of time spent in the alley during the 1-h test (emergence duration, ED).

Surgery

Two to 4 weeks following behavioral testing, the rats were anesthetized with an intraperitoneal (i.p.) injection of sodium pentobarbital (65 mg/kg) and mounted in a Kopf stereotaxic frame; the head position was adjusted to place bregma and lambda in the same horizontal plane. After retraction of the scalp, burr holes of approximately 2 mm diam were drilled bilaterally in the skull for the placement of stimulating and recording electrodes. The recording

electrode was implanted in the hilus of the dentate gyrus (3.3 mm posterior, 2.4 mm lateral, and 2.8–3.0 mm ventral to bregma) and the bipolar stimulating electrode in the medial perforant pathway (8.1 mm posterior, 4.4 mm lateral, and 2.5–4.0 mm ventral to bregma). The electrodes consisted of Epoxylite-coated stainless steel pins, with the recording and stimulating surfaces formed by removing the insulation at the tips (tip lengths = 50 and 500 μ m for the recording and stimulating electrodes, respectively). The ventral locations of both the recording and stimulating electrodes were adjusted to maximize the amplitude of the perforant-path evoked hilar responses. Body temperature was kept at approximately 37°C with a heating pad. Surgical anesthesia was maintained with booster injections (0.1 ml) of pentobarbital as needed.

Acute Electrophysiology

Electrophysiological testing [100- μ sec pulses delivered at 0.05 Hz; voltage adjusted to elicit an approximately 2-mV population spike (PS)] began after stable dentate hilar field potentials had been maintained for at least 30 min. The extracellular field potentials were amplified (gain = 100), bandpass filtered (1 Hz–10 kHz), digitized at 10 kHz, and written to disk on an AST Premium 386c computer (BrainWave Systems, Inc., Broomfield, CO). Following a 20-min baseline recording period, the subjects received the first of a series of five stimulation trains. The first stimulation train consisted of ten 40-msec, 25-Hz bursts delivered at the theta rhythm (5 Hz). The four remaining trains were delivered at 20-min intervals, in a graded series of intraburst frequencies (50, 100, 200, and 400 Hz) for the second through fifth trains, respectively. The first train was delivered at an intensity that elicited an approximately 1-mV PS prior to tetanization; the remaining trains were delivered at the same intensity. This series of stimulation trains permitted an evaluation of the threshold for LTP induction in each animal and increased the probability of evoking maximal LTP.

To assess the magnitude of LTP following each stimulation train, the percent change in PS amplitude and excitatory postsynaptic potential (EPSP) slope relative to the 20-min baseline was calculated for each 20-minute posttrain interval. In addition, input/output (I/O) functions consisting of five averaged field potentials at each of 10 different stimulation intensities were generated 20 min before the first stimulation train and 20 min following the last stimulation train. The I/O stimulation intensities for each animal were adjusted to elicit a range of field potentials; generally, the lowest intensity produced a pure population EPSP, whereas the highest intensity generated an asymptotic EPSP and PS.

Hormone Assays

A second group of 13 animals was concurrently tested under identical conditions. Two weeks following the emergence test, the animals were lightly anesthetized with methoxyflurane ($\approx$ 1 min) and decapitated. Trunk blood was collected in heparinized test tubes. The whole blood was centrifuged at 2,000 rpm for 20 min to isolate the plasma supernatant. Corticosterone radioimmunoassays were performed on the plasma samples by the Hormone Assay Core Laboratory, Population Research Center, Harbor-UCLA Medical Center, Los Angeles, CA. All of the physiological experiments were performed by an experimenter who was blind to the emergence performance of the animals.

RESULTS

Experiment 1

Electrophysiological recordings were made from 20 subjects 2 weeks after they had been tested in the emergence apparatus. Two subjects died during surgery, and

another 2 subjects exhibited unstable baseline evoked responses, leaving a total of 16 subjects to complete the experiment.

Emergence behavior. The mean ($\pm$ SEM) emergence latency for the 16 subjects was 18.6 ± 5.6 min (range = 2–60 min). The animals made a mean of 9.8 ± 2.4 entries (range = 0–31 entries) into the novel alley from their familiar nest box during the 1-h test. In addition, the animals spent an average of 8.3 ± 2.8 min (range = 0–39.3 min) in the novel alley, yielding a mean of 35.4 ± 9.4 sec (range = 0–112.1 sec) per alley entry.

Emergence neophobia and LTP threshold. Perforant-path evoked extracellular field potentials and the EPSP slope values generated from them during a representative 120-min recording session are shown in Figure 1. The mean percent changes ($\pm$ SEM) in EPSP slope (relative to baseline) following each stimulation train for all 16 subjects were as follows: 25 Hz = $-0.9 \pm 0.7\%$; 50 Hz = $-1.0 \pm 1.1\%$; 100 Hz = $5.2 \pm 1.9\%$; 200 Hz = $21.9 \pm 2.8\%$; 400 Hz = $32.3 \pm 3.8\%$. The mean percent changes in PS amplitude were as follows: 25 Hz = $3.7 \pm 8.5\%$; 50 Hz = $6.0 \pm 8.8\%$; 100 Hz = $112.0 \pm 37.3\%$; 200 Hz = $653.3 \pm 107.9\%$; 400 Hz = $1,426.7 \pm 375.1\%$. A one-way analysis of variance (ANOVA) with recording period (six levels) as a within-subject factor indicated that both the EPSP slope [$F(5,75) = 73.58, p < .001$] and the PS amplitude [$F(5,75) = 87.67, p < .001$] changed significantly during the recording session. Post hoc Fisher tests ($p < .01$) revealed that significant LTP of both the EPSP slope and the PS amplitude first occurred following the 100-Hz train and subsequently increased in magnitude following both the 200- and 400-Hz trains relative to the 20-min baseline. Thus, the initial appearance of LTP amounted to an increase relative to baseline of approximately 5% in EPSP slope and approximately 100% in PS amplitude.

To determine if individual differences in the threshold for LTP induction could be accounted for by emergence neophobia, the animals were divided into two groups ($n = 8$ per group) according to their emergence duration scores. Categorization of the subjects on the basis of their emergence duration scores revealed that 88% (7/8) of the animals in the neophobic group showed EPSP LTP (defined as a $\geq 5\%$ increase in EPSP slope) following the 100-Hz train, compared with only 25% (2/8) of the neophilic animals. All but one animal (a neophilic animal) exhibited significant EPSP LTP following the 200-Hz train. The lower threshold for LTP induction in neophobic subjects was significant [$\chi^2(1) = 2.8, p < .05$]. A similar pattern emerged for the incidence of PS LTP across stimulation sessions.

Emergence neophobia and LTP magnitude. The mean EPSP slope (A) and PS amplitude (B) LTP in neophobic and neophilic subjects are shown in Figure 2. A two-way ANOVA with factors of neophobia (two levels) and stimulation train (five levels) indicated that neophobic animals differed from neophilic animals in the magnitude

Figure 1. Percent change in excitatory postsynaptic potential (EPSP) slope during a 120-min recording session in a representative subject. Stimulation trains (frequency noted on the graph) were delivered at 20-min intervals. Note the appearance of long-term potentiation (LTP) following the 100-Hz train and the development of asymptotic LTP following the 400-Hz train. Traces at the top of the graph represent averaged perforant-path evoked extracellular field potentials recorded in the dentate gyrus during the 120-min recording session. Each field potential is an average of three evoked responses recorded during each 20-min period. Single-pulse stimulation of the perforant path (stimulation artifact appears as the initial negative-going spike) evoked a slow, positive-going population excitatory postsynaptic potential (EPSP). EPSP slope was measured as a change in voltage versus a fixed time interval on the rising phase of the positive-going slow potential. The population spike (PS) consisted of a fast, negative-going potential superimposed on the population EPSP. PS amplitude was measured as the voltage between a tangent drawn between PS onset and offset and the peak negativity of the PS. Note the robust potentiation of both the EPSP slope and PS amplitude following the 200- and 400-Hz trains. Calibration: sweep duration = 18 msec; peak EPSP amplitude = 11 mV.

of asymptotic EPSP LTP [$F(1,14) = 8.73, p < .01$], but not in the incidence of PS LTP [$F(1,14) = 2.25, p = .15$]. A significant interaction between neophobia and train frequency [$F(4,56) = 6.18, p < .001$] indicated that the difference in EPSP LTP found in neophobic and neophilic animals was expressed differentially across the stimulation sessions. Post hoc comparisons revealed that neophobic animals showed more EPSP LTP than did neophilic animals following both the 200-Hz ($p < .02$) and 400-Hz ($p < .01$) stimulation trains. There were no significant differences in the baseline I/O functions of neophobic and neophilic animals (data not shown).

Pearson correlations were calculated to determine if individual differences in emergence neophobia were predictive of the magnitude of LTP induced in each subject. As is apparent in Figure 3, both emergence duration (ED)

and emergence duration per alley entry (D/E) were significantly correlated with EPSP LTP [ED, $r = -.57, t(14) = 2.60, p < .05$; D/E, $r = -.74, t(14) = 4.15, p < .001$]. The correlations of emergence latency (EL) and emergence entries (ENT) with EPSP LTP did not reach statistical significance (EL, $r = .49$; ENT, $r = -.37$). In contrast, none of the emergence neophobia measures were significantly correlated with the magnitude of PS LTP (ED, $r = 0$; EL, $r = .08$; ENT, $r = .29$; D/E, $r = -.22$). It is not clear why we did not observe a correlation between emergence neophobia and PS LTP. In general, the high-frequency perforant-path stimulation used in the present study was associated with a much greater potentiation of the population spike than would be expected from the potentiation of the synaptic (EPSP) component alone, a phenomenon known as E-S potenti-

Figure 2. Mean ($\pm$ SEM) percent change in excitatory postsynaptic potential (EPSP) slope (A) and population spike (PS) amplitude (B) relative to baseline for neophobic ($n = 8$) and neophilic ($n = 8$) animals. Neophobic animals showed significantly more EPSP long-term potentiation (LTP) following the 200-Hz train ($*p < .02$) and the 400-Hz train ($**p < .01$). Although there was a trend for neophobic animals to show greater PS LTP than that showed by neophilic animals, this effect was not significant.

ation (Bliss & Lynch, 1988). Although E-S potentiation is common in the dentate gyrus in vivo, it may have been magnified in our experiments because of the relatively large number of stimulation pulses each animal received. Hence, it is possible that the pronounced EPSP-spike dissociation masked any correlation between PS LTP and emergence behavior.

Although the present data indicate a strong relationship between perforant-path LTP and neophobia, one could argue that this correlation was the result of long-term changes in synaptic efficacy that occurred in the hippo-

campus during emergence testing. In other words, changes in synaptic efficacy induced during the emergence testing may have occluded subsequent LTP induction. Three considerations argue against this interpretation: (1) Although changes in synaptic efficacy have been found to occur with exploration (Green et al., 1990; Sharp et al., 1989), they are relatively short-lived, typically lasting no more than 1 h after bouts of exploration. Any changes in synaptic efficacy that may have occurred during the emergence testing would probably have decayed 2 weeks after testing; (2) the emergence test is a relatively noninvasive pro-

Figure 3. Percent change in excitatory postsynaptic potential (EPSP) slope as a function of emergence duration (A), and emergence duration per alley entry (B) for 16 subjects. The correlation coefficients computed for the two data sets were highly significant (ED, $r = .57, p < .05$; D/E, $r = .74, p < .001$). Neophobic (non-emerging) animals tended to have greater EPSP slope long-term potentiation (LTP) than did neophilic (emerging) animals.

cedure that is considerably less traumatic or memorial than the multiple exposures to novel experiences and stimuli usually encountered by experimental animals procured from a distant supplier; and (3) the baseline I/O functions for the animals were essentially identical.

Experiment 2

To examine the possibility that plasma corticosterone levels contributed to the correlation that we observed between LTP and emergence behavior, an additional 13 subjects were run in the emergence apparatus. Trunk blood was collected from these subjects 2 weeks after emergence testing in order to assess basal plasma corticosterone levels.

Emergence behavior. The emergence behavior in these animals was comparable to that observed in the first set of animals. The mean ($\pm$ SEM) emergence latency for the 13 subjects was 4.4 ± 0.5 min (range = 2.7 - 8.2 min). During the 1-h test, the animals made a mean of 20.1 ± 0.3 entries (range = 10-46 entries) into the novel alley from their familiar nest box. In addition, the animals spent an average of 23.9 ± 2.7 min (range = 8.9-41.1 min) in the novel alley, yielding a mean of 84.2 ± 14.2 sec (range = 34.6-205.5 sec) per alley entry.

Corticosterone levels. Corticosterone radioimmunoassays revealed that the plasma corticosterone levels in these subjects were in the "normal," nonstressed range [0.5 - 13.6 μ g/dl; mean ($\pm$ SEM) = 5.7 ± 1.3 μ g/dl]. As illustrated in Figure 4, a significant correlation was found between plasma corticosterone levels and emergence duration [$r = .65$, $t(11) = 2.85$, $p < .02$]. None of the other measures of emergence behavior correlated with

plasma corticosterone levels (data not shown). Thus, animals that tended to emerge into the novel alley had higher basal plasma corticosterone levels than those that remained in the familiar compartment. Although we did not measure plasma corticosterone levels in animals during the emergence testing, a recent report (Misslin & Cigrang, 1986) indicates that mice given a choice between novel and familiar alternatives in an exploratory task do not show stress-related elevations in plasma corticosterone levels. Stress-induced elevations in corticosterone levels occurred only when animals were forcibly exposed to a novel situation. Therefore, we assume that the basal plasma corticosterone levels measured 2 weeks after emergence testing were compatible with the levels during emergence testing.

We were somewhat surprised to find that neophobic animals had relatively lower plasma corticosterone levels than did neophilic animals because of the previously cited evidence that both acute and chronic stressors markedly enhance neophobia. The present data would suggest that basal corticosterone levels may modulate neotic behavior in a somewhat different manner from that which occurs under stressful conditions. This pattern of results might be accounted for by the differential occupation of Type I (high affinity) and Type II (low affinity) glucocorticoid receptors under stressed and nonstressed conditions (Joels & de Kloet, 1992). Type I receptors are located primarily in the hippocampus and other limbic structures, whereas Type II receptors are more widely distributed throughout the brain (Magariños, Ferrini, & De Nicola, 1989; Reul & de Kloet, 1985). It has been suggested that saturation of Type I receptors under nonstressed conditions (serum corticosterone levels of 1-10 μ g/dl) mediates a tonic influence of corticosterone on limbic structures, a process that is relevant to the threshold of the stress response and the organization of behavioral responses to stress (Joels & de Kloet, 1992). In contrast, Type II receptors may play a more direct role in information storage under conditions in which corticosterone exceeds basal levels (serum levels of 20-100 μ g/dl). In this regard, it is tempting to speculate that behavioral responses to novelty under nonstressed conditions are mediated primarily by Type I receptors, and are fundamentally different from Type II mediated responses.

DISCUSSION

The results of the present study demonstrate that neophobic animals have a significantly lower threshold for induction of perforant-path granule-cell LTP than do neophilic animals. Moreover, neophobic animals exhibited significantly more EPSP LTP than did their neophilic counterparts. It is plausible that these results indicate that LTP and neophobia are indirectly linked through a habituation mechanism. Neotic choice behavior is usually found to be more robust in situations offering a choice between relatively more dichotomous novel and familiar alternatives than between relatively less dichotomous al-

Figure 4. The relationship between emergence duration (in minutes) and plasma corticosterone (μ g/dl) in 13 animals. A Pearson correlation coefficient computed for the data set was highly significant ($r = .65$, $p < .02$). Neophobic (non-emerging) animals tended to have lower circulating levels of corticosterone than did neophilic (emerging) animals.

ternatives. For example, Mitchell, Kirschbaum, and Perry (1975) found that rats were more likely to avoid a complex of novel stimuli in a more familiar environment than in a relatively more novel environment. Applied to the present results, this view suggests that the rats with a low threshold to induce LTP and/or high levels of LTP efficiently habituated to the initially novel nest box. When subsequently tested for emergence behavior, they showed a neophobic reluctance to emerge because, for them, there was a clear distinction between the now familiar nest box and the novel alley. Conversely, the animals with a high threshold for LTP induction and/or low levels of LTP were inefficient at habituating to the nest box. For these animals, the distinction between the novel and familiar alternatives was relatively unclear, resulting in reduced neophobia during emergence testing.

Another plausible interpretation for the present data can be drawn from theories that attribute motor functions to the hippocampus (Blanchard, Blanchard, Lee, & Fukunaga, 1977; Douglas, 1967; Vanderwolf, 1969). Because animals that emerge into a novel environment are necessarily more active than non-emerging animals, one might be tempted to attribute the correlation between emergence behavior and LTP to individual differences in activity levels. However, emergence neophobia can be used to predict behavior in other neotic choice paradigms that do not have a prominent motoric component. For example, emergence neophobia is correlated with long-delay conditioned taste aversions to saccharin (Mitchell, 1985). Because there is no difference in the motor activity expended when drinking from a bottle containing a novel saccharin solution or an identical bottle containing familiar tap water, these results cannot be attributed to differences in motor activity.

Another, perhaps more relevant example, comes from recent data indicating that animals that are more active in an emergence test are actually less active in a contrafreeloading test (Mitchell, Wong, & Yang, 1993). Contrafreeloading is a neotic operant task in which a rat's neophobic reluctance to eat from a novel food source competes against its reluctance to expend energy needlessly (Mitchell, Becnel, & Blue, 1981; Mitchell, Fish, & Calica, 1982; Mitchell, Scott, & Williams, 1973). In this task, animals are first trained to barpress for food pellets and then are presented with a choice between this now familiar "earned" food source and a novel source consisting of a food cup filled with identical, but more accessible, "free" pellets. Non-emerging neophobic animals that are inactive during an emergence test continue to actively barpress in the presence of free food. Conversely, emerging neophilic animals are more active in an emergence test but less active in the contrafreeloading task—they rapidly shift to the free food. Hence, neophobia, not activity, appears to dominate choice behavior in neotic choice tasks.

The correlation between emergence behavior and plasma corticosterone levels suggests a possible mechanism for the differences in EPSP LTP observed between neophobic and neophilic animals. In the present study,

neophobic animals tended to have both lower plasma corticosterone levels and elevated perforant-path LTP. This finding agrees with recent reports indicating that glucocorticoid hormones have an inhibitory effect on LTP induction in area CA1 in vivo (Bennett et al., 1991; Pavlides, Watanabe, & McEwen, 1991). Two points deserve mention, however: First, the corticosterone levels reported in the Bennett et al. (1991) study were in the stress range (20–100 $\mu\text{g/dl}$) as a consequence of urethane anesthesia. In contrast, the levels of corticosterone measured in the present study were all within the basal, nonstressed range (0.5–13.6 $\mu\text{g/dl}$). This suggests that corticosterone may have a modulatory influence on hippocampal plasticity in both stressed and non-stressed animals. However, it must be noted that corticosterone levels do not have a direct role in the impairment of LTP with high stress (Shors, Foy, Levine, & Thompson, 1990; Shors, Levine, & Thompson, 1990). Second, Bennett et al. (1991) reported a suppression of PS LTP with elevated corticosterone levels, whereas the results of the present study indicate no relationship between emergence behavior and, by inference, corticosterone levels on this measure. As was suggested earlier, however, one reason for this discrepancy may be the massive E-S potentiation we observe in our LTP preparation, a phenomenon that may be less likely to occur in the primed-burst paradigm used by Bennett et al. (1991).

The differential induction of perforant-path LTP in neophobic and neophilic animals may reflect individual differences in hippocampal glutamate receptor populations. In area CA1 and the dentate gyrus, LTP induction requires activation of *N*-methyl-D-aspartate (NMDA) receptors, a subclass of glutamate receptors (Collingridge, Kehl, & McLennan, 1983; Maren, Baudry, & Thompson, 1991, 1992; Morris et al., 1986). Once induced, the expression of LTP is maintained by a selective modification of post-synaptic α -amino-3-hydroxy-5-methyl-4-isoxazolepropionate (AMPA) receptors, a non-NMDA subclass of glutamate receptors (Foster & McNaughton, 1991; Lynch & Baudry, 1991; Maren, Baudry, & Thompson, 1992; Tocco, Maren, Shors, Baudry, & Thompson, 1992). Recently, it has been reported that rats bred for superior learning in a shuttlebox avoidance task exhibit greater perforant-path LTP and an increased number of hippocampal NMDA receptors than do poor learners (Keller, Borghese, Carrer, & Ramirez, in press; Ramirez & Carrer, 1989; Ramirez, Orsingher, & Carrer, 1988). Moreover, exogenous corticosterone application is known to decrease AMPA receptor binding in the hippocampus (Tocco, Shors, Standley, Baudry, & Thompson, 1991), indicating that corticosterone may exert its effect on LTP mechanisms via an interaction with glutamate receptors. These results suggest the interesting possibility that individual differences in neophobia and LTP induction are related in some way to properties of hippocampal glutamate receptors. We are currently pursuing experiments to test this prediction.

In conclusion, we have demonstrated that emergence behavior in an exploratory task with a clear distinction

between novel and familiar alternatives predicts the threshold for and the asymptotic level of synaptic LTP in the dentate gyrus in vivo. Nonexploratory (neophobic) animals had a lower threshold for LTP induction and showed significantly greater LTP than did their exploratory (neophilic) counterparts. The correlations between emergence behavior and LTP on the one hand and plasma corticosterone levels on the other suggest that all three share a common mechanism. We speculate that endogenous levels of LTP and/or the capacity to exhibit LTP in vivo may modulate neotic choice behavior through an interaction of glucocorticoid hormones and habituation.

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