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BRIEF COMMUNICATION

Learned Helplessness Induction Decreases *in Vivo* Cortical Serotonin Release

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PETTY, F. AND A. D. SHERMAN. *Learned helplessness induction decreases* in vivo *cortical serotonin release.* PHARMACOL BIOCHEM BEHAV **18**(4) 649–650, 1983.—Frontal neocortices of freely moving, unanesthetized rats were perfused before, during, and after exposure to 40 min of uncued pulsed random footshock. Animals developing nontransient learned helplessness had lower levels of serotonin in cortical perfusate than those failing to develop helplessness.

Learned helplessness Serotonin release Footshock

THE frontal neocortex seems to play a unique role in the reversal of learned helplessness by antidepressant drugs, with serotonergic mechanisms mediating drug action in this animal model of depression. When tricyclic antidepressant drugs were injected into 9 different brain structures, a behavioral reversal of helplessness was found only following injection into the frontal neocortex. This reversal was also seen after injection of serotonin into this region but not norepinephrine, GABA, acetylcholine, glutamate, or dopamine [1].

In vitro data also support the role of serotonin as mediating helplessness reversal by antidepressant drugs. Helpless rats have decreased calcium-specific release of serotonin from cortical slices compared to controls, while chronic treatment with tricyclic antidepressants increases calcium specific serotonin release. These effects were additive, with animals receiving both helplessness induction and chronic tricyclic treatment having serotonin release essentially in the normal range, as well as normal escape performance behavior. These effects were not seen with norepinephrine [2].

We were interested as to whether similar effects might be seen *in vivo* during exposure of rats to uncontrollable footshock.

METHOD

Male 200–250 g Sprague-Dawley derived rats were used in all experiments. Animals were singly housed with free access to food and water.

After administration of surgical anesthesia (chloral hydrate 400 mg/kg), animals were mounted in a stereotaxic apparatus (Stoelting-Stellar). The skull was exposed with an incision extending from the interobital line to lambda. A burr hole was drilled above frontal neocortex using coordinates of Pellegrino *et al.* [3] (from bregma 3 mm anterior and 2 mm lateral). After removal of dura, a push/pull cannula assembly was inserted (Plastic Products, designed according to Myers [4]) into cortex such that the pull cannula rested 0.3 mm below cortical surface. Perfusion was then established (Harvard apparatus reciprocal syringe pump) with artificial cerebral spinal fluid [5]. The guide cannula was then cemented to skull by means of 3 0-80 stainless steel set screws and dental acrylic, with a dummy cannula replacing the push cannula.

Animals were allowed 7 days for recovery from surgery and perfusion was then reestablished under light ether anesthesia. Animals were then placed in an aluminum modular shockbox with stainless steel grid floor and four 10 min baseline samples of perfusate collected. Animals were then subjected to our standard learned helplessness induction procedure [1] which consists of a random pulsed low-intensity (0.8 mA) footshock and an additional four 10 min aliquots of perfusate were collected during this time. After footshock offset another four 10 min aliquots were collected.

For purposes of analysis, duplicate samples were pooled such that assays were performed on 2 baseline samples, 2 during helplessness induction and 2 after helplessness induction. Serotonin was determined by high performance liquid chromatography using electrochemical detection [6]. In all cases in which the flow of perfusate varied or air bubbles or blood were observed in the pull lines, animals were discarded from the experiment. Results were calculated as a percent of each animal's control value.

RESULTS

There was considerable variability in the samples obtained during helplessness induction (Fig. 1). This may reflect the fact that the animals were receiving footshock dur-

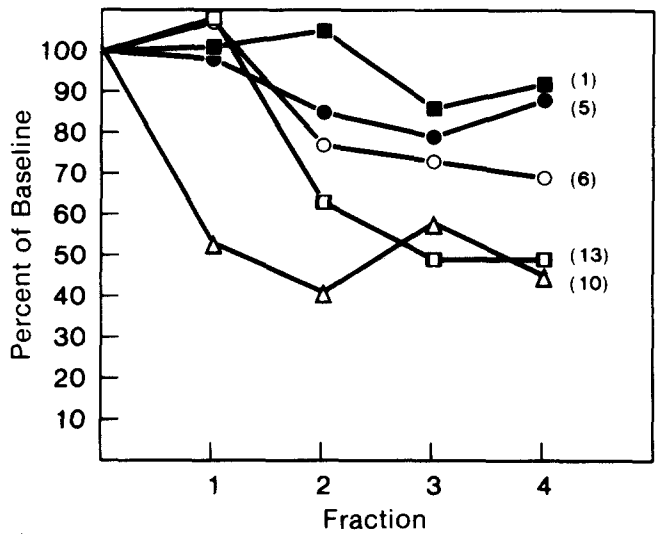

FIG. 1. Serotonin release from cortex of rats undergoing learned helplessness induction expressed as a percent of baseline serotonin release. Baseline values were 3.54±1.42 pmol/μl of perfusate. Fractions 1 and 2 were collected during exposure to inescapable shock, fractions 3 and 4 after shock termination. Each curve represents one animal. Black symbols correspond to animals which did not develop non-transient (24 hours) learned helplessness. Open symbols correspond to animals which developed helplessness. Numbers in brackets are the number of escape failures during 15 trials of a bar-press escape task. Naive controls score 1–5 escape failures, helpless rats score 6–15 failures [1].

ing this time and were often exhibiting considerable motor arousal. However, after learned helplessness induction, clear differences could be demonstrated between animals who remained helpless when tested at 24 hours and those in whom helplessness dissipated during this period of time. In summary, this preliminary experiment confirms our findings *in vitro* of decreased serotonergic activity in cortex being related to learned helplessness induction and additionally demonstrates the feasibility of *in vivo* push/pull perfusion techniques for studying rats during aversive behavioral conditioning.

ACKNOWLEDGEMENTS

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