Pharmacology Biochemistry & Behavior, Vol. 30, pp. 787–789. © Pergamon Press plc, 1988. Printed in the U.S.A. 0091-3057/88 $3.00 + .00

BRIEF COMMUNICATION

Conditioned Place Preference Produced by Intra-Hippocampal Morphine[1]

WILLIAM A. CORRIGALL AND MARY ANN LINSEMAN

Neurobiology Section, Addiction Research Foundation
33 Russell Street, Toronto, Ontario, M5S 2S1, Canada

Received 22 September 1987

CORRIGALL, W. A. AND M. A. LINSEMAN. *Conditioned place preference produced by intra-hippocampal morphine.* PHARMACOL BIOCHEM BEHAV 30(3) 787–789, 1988.—Unilateral microinjections of morphine sulphate into the rat hippocampus were found to produce a conditioned place preference, whereas equi-volume saline injections into the same region in a separate control group were without effect. The results are discussed in terms of possible reward or habituation functions of the hippocampus.

Hippocampus Conditioned place preference Morphine Habituation Opiate Reinforcement

ELECTROPHYSIOLOGICAL studies have shown that opiate agonists have an excitatory effect within the hippocampus, whether measured in field potential activity (e.g., [6,9]) or in single neuron responses (e.g., [7, 8, 16]). Receptor binding studies have shown that presumptive mu-type binding sites are distributed throughout the pyramidal cell layer (see review in [4]), and there has been some corroboration electrophysiologically that it is a mu- and/or delta-receptor population which mediates the excitation [1, 13, 14].

In spite of the clear evidence for the electrophysiological effect, however, there have been no studies designed to examine behavioral consequences of agonist treatment in this region of the hippocampus. It is therefore not known what the manifestation of opiate action on hippocampal neurons is, if any, at a higher level of integration. For this reason, we examined the ability of microinjections of morphine into the hippocampal region to produce several non-conditioned behaviors, but found no effects of doses up to 50 micrograms on rectal temperature, analgesia or catalepsy (unpublished data). Because of the purported role of the hippocampus in learning and memory functions, we subsequently tested the ability of intra-hippocampal morphine infusions to produce a conditioned place preference, the results of which are described below.

METHOD

Subjects for these experiments were 12 male Sprague-Dawley rats (Charles River; Lachine, Que), 350–450 g in weight. After habituation to our animal colony they were prepared with unilateral brain cannulae (Plastic Products, Roanoke, VA). Cannula guides were cut to reach a position in the brain just dorsal to the CA1 region of the hippocampus, and were implanted stereotaxically under pentobarbital anaesthesia according to the following coordinates: A-P: −3 mm/L: 1.5 mm relative to bregma and perpendicular to the skull. Except during drug injection, guide cannulae were kept plugged with obturators.

A week following surgery animals were given a 15-minute pretest on each of 3 successive days; during this period, the amount of time spent by the animal in each section of a 3-compartment chamber was measured. The chamber was similar to that used by others [10] and consisted of a central start area with grey plywood floor and walls, and two distinctive environments differing from each other only in floor covering, one being black Plexiglas and the other wooden chips over a grid. The walls of these environments were matte black in color. Following pretests, animals were randomly assigned to two groups. Animals of each group were administered either saline or 20 micrograms of morphine respectively and confined singly for 30 minutes to their non-preferred environment. (The 20 microgram dose was chosen since we earlier observed that the morphine dose threshold for affecting hippocampal neuronal activity was relatively high both in vivo [9] and in vitro [6].) Infusions were done through cannulae extending 0.5 mm beyond the guides and were performed slowly over approximately two minutes by means of a gas-tight microsyringe advanced by a manually-driven micrometer screw; drug delivery was confirmed by watching the advancement of a small air bubble in the polyethylene line carrying drug solution to the cannula. Pairing of drug or saline with the environment was carried out once each day for four days. On the fifth day a test session was conducted in which the animals had access to the full test box for 15 minutes.

[1]The views expressed in this publication are those of the authors and do not necessarily reflect those of the Addiction Research Foundation.

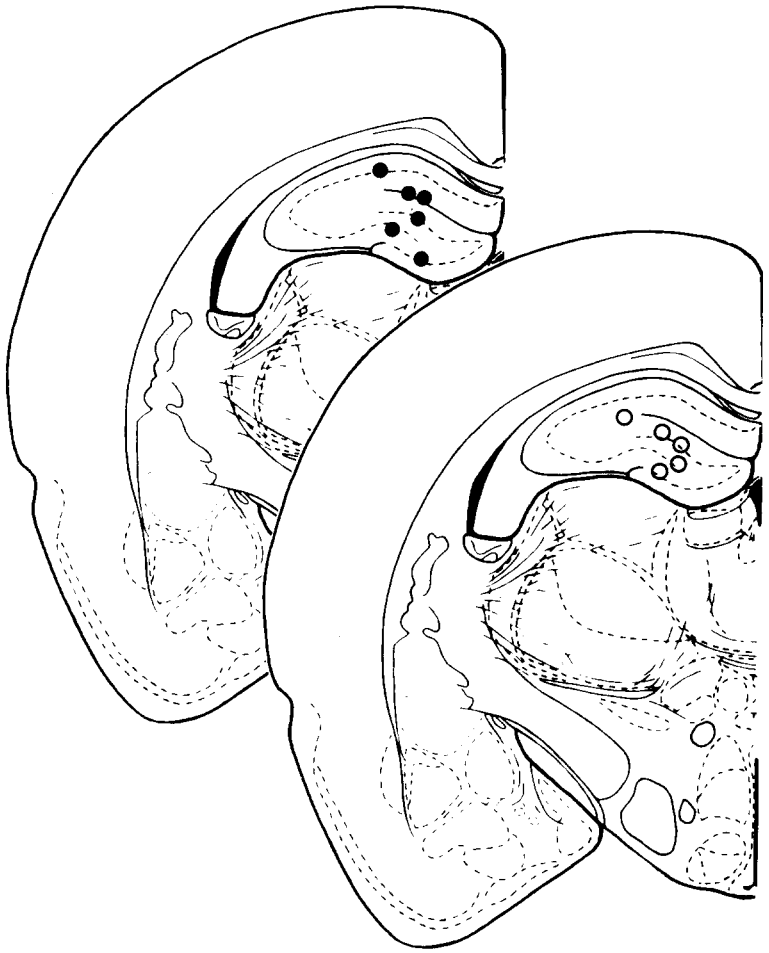

FIG. 1. Schematic representation of the location of cannula tips in the hippocampal function. Filled circles represent the histology for the group paired with morphine; open circles represent the animals paired with saline.

At the conclusion of the experiment, animals were given an overdose of pentobarbital, and perfused transcardially with saline followed by 10% formalin. Brains were removed, and following further fixation and dehydration, were sliced into 50 micrometer sections which were mounted for histological determination of the cannula tips. All animals were found to have cannula placements within the hippocampus (see Fig. 1), except for one of the saline control animals whose cannula tip could not be identified.

Differences between pre- and posttreatment scores were analyzed using a t-test for independent groups.

RESULTS AND DISCUSSION

The results of the place preference tests are shown in Fig. 2. After treatment animals which received morphine spent more time in the drug-paired environment than saline-treated animals spent in the saline-treated environment, $t(10)=3.63$, $p<0.05$.

The usual interpretation of place preference data has been that the drug-paired environment has assumed some secondary rewarding property as a consequence of its being regularly paired with the positive affective properties of the drug (e.g., see [10]), although it should be noted that a relationship between place conditioning and drug reinforcement has not been established. In this context, our findings suggest that opiate receptors in the vicinity of the microinjection sites in hippocampus are involved in some feature of drug reward, along with a variety of other brain loci [5, 11, 15]. The observation that naloxone reduces hippocampus self-stimulation would also support a role for hippocampal opiates in reinforcement [2].

However, while it is tempting to adopt an interpretation in which hippocampal neurons play a role in opiate reward, one must be cautious in view of alternative explanations. For example, Scoles and Siegel [12] have recently demonstrated the role played by saline trials in conditioned place preference, and have suggested that place preference conditioning with opiates could be due to an opiate-produced attenuation of habituation. Although the findings of Scoles and Siegel differ in form from those reported here (i.e., the Scoles and Siegel data show no change in preference for the morphine paired environment and a decrease in preference for that paired with saline), they may be important to the interpreta-

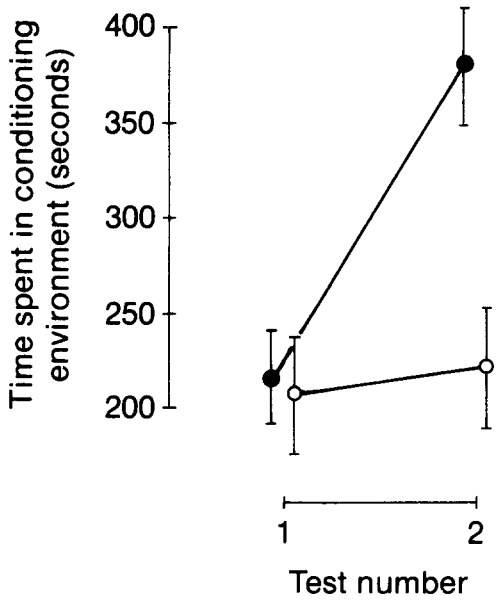

FIG. 2. Results of test trials before and after repetitive pairings of infusions of either morphine (20 micrograms) or saline with the less preferred environment. Data is presented as the mean and standard error of the mean (n=6 at each point). ○=Saline; ●=Morphine.

tion of the present hippocampal data. Also, it has been shown that hippocampal opiates may impair recent memory formation [3]. It is not unreasonable, therefore, to speculate that within the place conditioning procedure, intrahippocampal morphine might result in impaired habituation to, or memory of, the drug-paired environment as compared to the saline environment. On balance, then, in the test situation, the drug-paired environment would be the more novel, resulting in increased exploration of it and thereby in an apparent increased preference for it. Future experiments to test between the reward or habituation interpretation of the data reported here might profitably examine whether hippocampal receptors regulate the self-administration of opiate agonists, as has been done for other brain areas [5].

ACKNOWLEDGEMENTS

This research was supported by the Addiction Research Foundation of Ontario. We are grateful to Myrnalee Elliott and RoseMarie D'Onofrio for help with these experiments.

REFERENCES

1. Bostock, E., Dingledine, R.; Xu, G.; Cheng, K-J. Mu-opioid receptors participate in the excitatory effect of opiates in the hippocampal slice. J. Pharmacol. Exp. Ther. 231:512–517; 1984.
2. Collier, T. J.; Routtenberg, A. Electrical self-stimulation of dentate gyrus granule cells. Behav. Neural. Biol. 42:85–90; 1984.
3. Collier, T. J.; Routtenberg, A. Selective impairment of declarative memory following stimulation of dentate gyrus granule cells: a naloxone-sensitive effect. Brain Res. 310:384–387; 1984.
4. Corrigall, W. A. Opiates and the hippocampus: A review of the functional and morphological evidence. Pharmacol. Biochem. Behav. 18:255–262; 1983.
5. Corrigall, W. A. Heroin self-administration: Effects of antagonist treatment in lateral hypothalamus. Pharmacol. Biochem. Behav. 27:693–700; 1987.
6. Corrigall, W. A.; Linseman, M. A. A specific effect of morphine on evoked activity in the rat hippocampal slice. Brain Res. 192:227–238; 1980.
7. Dunwiddie, T.; Mueller, A.; Palmer, M.; Stewart, J.; Hoffer, B. Electrophysiological interactions of enkephalins with neuronal circuitry in the rat hippocampus. I. Effects on pyramidal cell activity. Brain Res. 184:311–330; 1980.
8. Gähwiler, B. H.; Maurer, R. Involvement of μ-receptors in the opioid-induced generation of bursting discharges in hippocampal pyramidal cells. Regul. Pept. 2:91–96; 1981.
9. Linseman, M. A.; Corrigall, W. A. Effects of morphine on CA1 versus dentate field potentials following systemic administration in freely-moving rats. Neuropharmacology 21:361–366; 1982.
10. Mucha, R. F.; Van der Kooy, D.; O'Shaughnessy, M.; Bucenieks, P. Drug reinforcement studied by the use of place conditioning in the rat. Brain Res. 243:91–105; 1982.
11. Phillips, A. G.; Le Piane, F. G. Reinforcing effects of morphine microinjections into the ventral tegmental area. Pharmacol. Biochem. Behav. 12:965–968; 1980.
12. Scoles, M. T.; Siegel, S. A potential role of saline trials in morphine-induced place-preference conditioning. Pharmacol. Biochem. Behav. 25:1169–1173; 1986.
13. Valentino, R. J.; Bostock, E.; Dingledine, R. Opioid pharmacology in the rat hippocampal slice. Life Sci. 31:2339–2342; 1982.
14. Valentino, R. J.; Dingledine, R. Pharmacological characterization of opioid effects in the rat hippocampal slice. J. Pharmacol. Exp. Ther. 223:502–509; 1982.
15. Van der Kooy, D.; Mucha, R. F.; O'Shaughnessy, M.; Bucenieks, P. Reinforcing effects of brain microinjections of morphine revealed by conditioned place preference. Brain Res. 243:107–117; 1982.
16. Zieglgänsberger, W.; French, E. D.; Siggins, G. R.; Bloom, F. E. Opioid peptides may excite hippocampal pyramidal neurons by inhibiting adjacent interneurons. Science 205:415–417; 1979.