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Effects of Parenteral Morphine and Oral Methadone on Self-Stimulation in the Rat[1]

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PRESHAW, R. L., H. ZENICK AND R. M. STUTZ. *Effects of parenteral morphine and oral methadone on self-stimulation in the rat.* PHARMAC. BIOCHEM. BEHAV. **16**(1) 81–85, 1982.—Facilitation of self-stimulation has been reported following the administration of various opiates. Methadone, a synthetic narcotic used in the treatment of narcotic addiction, has recently been demonstrated to facilitate self-stimulation when administered parenterally. The present study examined the effects of orally administered methadone (20 and 30 mg/kg), the route of administration used clinically, on MFB-LH self-stimulation at 2.5, 5, 8, 12, 17, and 24 hours post-administration. Reliable facilitation was observed at 2.5 hours post-administration. However, the effect of methadone was less pronounced than that observed with a dose of parenteral morphine which was apporximately equivalent in terms of analgesic potency.

Methadone Morphine Self-stimulation Euphoria Opiates

IT has been suggested that investigating the effects of potentially addictive drugs (e.g., morphine, cocaine, amphetamine) on self-stimulation (SS) may clarify the mechanisms underlying the reinforcing properties of these drugs [4, 16, 17]. Drugs with high addiction liability generally facilitate SS, whereas drugs with little or no addiction liability produce little or no facilitation [23].

Facilitation of SS has been reported following the administration of various opiates [1, 3, 8, 12, 14, 22, 25, 28]. Methadone, a synthetic opiate, is of particular interest since a widely used treatment for human narcotic addiction consists of maintaining addicts on orally administered methadone. In the animal literature it has been unclear whether methadone facilitates SS. Pert and Hulsebus [22] reported facilitation of SS by parenteral methadone (no dosage reported) but Pert [21], employing doses of 0.1–3.0 mg/kg, and Schaefer and Holtzman [24], using doses of 0.1–3.0 mg/kg, did not observe facilitation of SS by parenteral methadone. However, using a larger drug dose (10 mg/kg) and a wider range of testing times, Stutz, Maroli, Tsang and Harvan [25] demonstrated facilitation of SS by parenteral methadone.

No studies have been reported examining the effect of orally administered methadone on SS. Yet this route of administration is used clinically and has marked differences relative to the parenteral route [10]. The present study was designed to examine the effect of oral methadone on SS at various post-administration times.

EXPERIMENT 1

Experiment 1 was designed to identify opiate facilitators (as defined by individual SS rates under morphine relative to their responding under saline) in order to subsequently examine their responsiveness to methadone. Large and reliable individual differences in SS behavior occur in response to opiates [23,26]. The reason for these differences has not been determined but may represent variations in individual reactivity to the drug itself (as has been reported for hedonic value in humans [19]). Individual differences may also result from slight variations in electrode locus [15]. In any event, only those animals classified as opiate-facilitators in Experiment 1 were tested in Experiment 2.

In addition, the results of Experiment 1 permitted com-

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parison of the effects of methadone (obtained in Experiment 2) with those of the prototypic opiate, morphine, using the same subjects and experimental parameters.

METHOD

Animals

The animals were naive male Sprague-Dawley albino rats born and reared in the animal colony maintained in the Department of Psychology at the University of Cincinnati. At the time of surgery, the animals weighed between 350–450 g. Thirty-seven of the animals implanted with intracranial electrodes were included in the experiment. Between experimental sessions animals were housed individually in a colony room which was artificially illuminated between 6:00 a.m. and 6:00 p.m. Food and water were available only between 3:30 p.m. and 5:00 p.m. throughout all experiments in order to reduced variability in rate of drug absorption as a function of stomach load. Drug administration always occurred at 9:00 a.m.

Surgery

Each animal was stereotactically implanted with a bipolar stimulating electrode (Plastic Products Co., MS-303-.018-.312-.010) under sodium pentobarbital (Nembutal) anesthesia (45 mg/kg, IP). Electrodes were aimed at the postero-lateral hypothalamus/medial forebrain bundle. With the skull horizontal between bregma and lambda, the coordinates were 4.5 mm posterior to bregma, 1.5 mm lateral to the mid-line, and 8.5 mm below the surface of the skull. This corresponds to the de Groot [6] coordinates: AP 5.0, V −2.75, L 1.75.

Procedure

After at least one week for recovery from surgery, daily 30 min sessions were given in which animals were trained to SS and rates were allowed to stabilize. Each depression of the lever resulted in the delivery of a 300 msec train of 60 Hz sine waves through a constant current circuit. During this period, a current intensity (never greater than 50 μA rms) was selected for each animal which yielded stable response rates of at least 300 barpresses per 10 min. This current intensity remained constant for a given animal throughout all experiments. Each SS session was 12 min in length. The first 2 min were considered a warm-up period during which the animals were primed if they did not immediately begin to SS, although priming was rarely necessary. The number of barpresses was recorded by an electromechanical counter during the last 10 min of the session.

For each treatment condition, animals were treated for five consecutive days and tested for the last three days of treatment. The first two daily injections for a treatment were to allow tolerance to occur to the rate suppressive effects of the particular drug and dosage. All animals received each treatment condition in the following order: SAL (normal saline, IP), MO-10 (10 mg/kg of morphine, IP), and MO-15 (15 mg/kg of morphine, IP). The doses of morphine were calculated in terms of the salt and the volume of injection was 1 ml/kg body weight. All animals were tested 2 and 4 hours post-administration.

RESULTS AND DISCUSSION

A three within factors ANOVA (Treatment, Days and Time post-injection) yielded significant effects for Treatment, $F(1,36)=13.92$, $p<0.01$, Time, $F(1,36)=7.62$, $p<0.01$, and the Day×Treatment interaction, $F(1,36)=5.45$, $p<0.05$. The significant effect for treatment represents a dose related increase in responding following morphine administration (see Fig. 1).

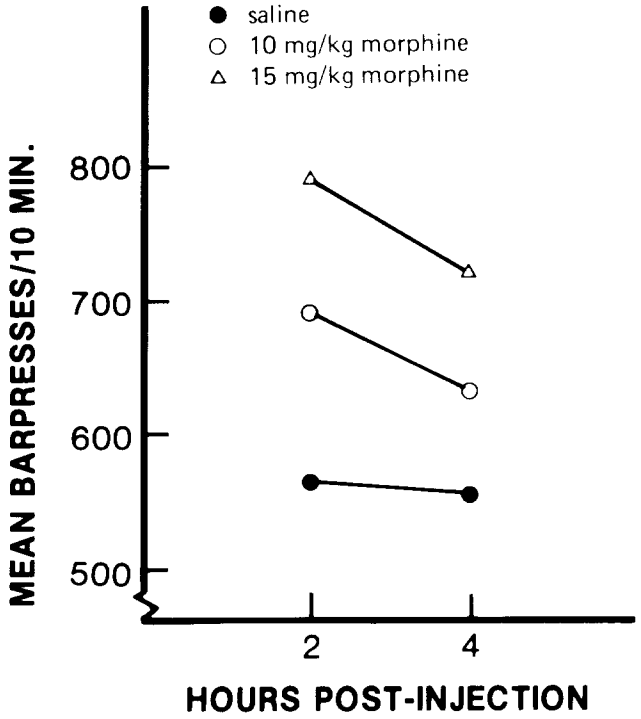

FIG. 1. Self-stimulation rates following parenteral administration of saline, 10 mg/kg or 15 mg/kg of morphine (data collapsed over days). Data presented are from the same animals as those representing the Drug Group of Experiment 2.

Paired *t*-tests (data collapsed across days) yielded a significant difference between SAL and MO-10, SAL and MO-15, and MO-10 and MO-15 treatments. The facilitation of SS observed following parenteral morphine replicates the findings of several other investigators [1, 3, 17]. As previously reported [23,26], large and reliable individual differences in SS rates occurred in response to morphine. The SS rates of 32 of the 37 animals under morphine treatment exceeded their mean response rate during saline testing, and were considered opiate-facilitators. The 5 remaining animals were not included in Experiment 2.

The Day×Treatment interaction was due to an increase in responding over days during morphine treatment as compared to saline treatment. This suggests the possibility that tolerance was still occurring to the rate suppressive effects of morphine at this point. Thus four daily administrations of the test drug preceded SS testing in Experiment 2.

EXPERIMENT 2

The purpose of Experiment 2 was to examine the time course of the effects of oral methadone on SS.

METHOD

Animals

The 32 animals classified as opiate-facilitators in Experiment 1 were divided into groups matched on the degree of facilitation observed during morphine testing (i.e., the number of standard deviations above saline responding). One group was designated to receive methadone treatment (Drug Group), the other served as a saline control (Saline Group). Three animals in each group did not complete Experiment 2 due to loss of the electrode assembly or death of unknown causes.

Procedure

The procedure was similar to that of Experiment 1 with the following exceptions. The treatment conditions for the Drug Group consisted of SAL (normal saline, PO), ME-20 (20 mg/kg of methadone, PO), and ME-30 (30 mg/kg of methadone, PO). The treatment conditions for the Saline Group always consisted of normal saline (PO). Animals were treated for seven consecutive days and tested for the last three days of treatment. All animals were tested 2.5, 5, 8, 12, 17 and 24 hours post-intubation. This experiment began 5 days following Experiment 1. The doses of methadone HCl (Lilly) were calculated in terms of the salt and the volume of intubation was 1 ml/kg body weight. Morphine and methadone are approximately equally potent when administered parenterally and methadone administered orally is approximately half as potent as when administered parenterally [2,18]. The 20 mg/kg dose of oral methadone was chosen to be approximately equally potent as the 10 mg/kg dose of parenteral methadone previously shown to facilitate SS in this laboratory [25]. Pilot work using doses substantially higher than 30 mg/kg were found to produce a high degree of muscular rigidity which did not show complete tolerance by the fourth daily administration. The high and low doses of morphine (Experiment 1) were selected to be about equally potent to the high and low doses of methadone (Experiment 2).

Finally, in order to determine whether the experience with methadone altered the animals' responsiveness to morphine, the effects of 15 mg/kg of morphine on SS were re-examined eight days following the ME-30 treatment. The procedure was identical to that of MO-15 in Experiment 1.

Histology

Animals were sacrificed with an overdose of sodium pentobarbital and perfused intracardially with physiological saline followed by 10% Formalin. The frozen brains were sliced in 40 μm sections and microscopically examined to determine the locations of the electrode tips.

RESULTS

Percent change from SAL scores for each animal were calculated and analyzed using a one between (Groups), two within (Treatments and Time) factors ANOVA. This analysis yielded significant effects of Time, $F(1,22)=9.33$, $p<0.01$, and Group×Time interaction, $F(1,22)=7.74$, $p<0.025$. Both groups exhibited a similar pattern of responding over time during SAL treatment (shown for the Drug Group in Fig. 2). A circadian effect has previously been demonstrated in rats pressing for rewarding ESB [5,27]. It has also been reported that food deprivation can increase SS rates [9,20]. It is likely, then, that some combination of these factors produced the observed pattern of responding over time.

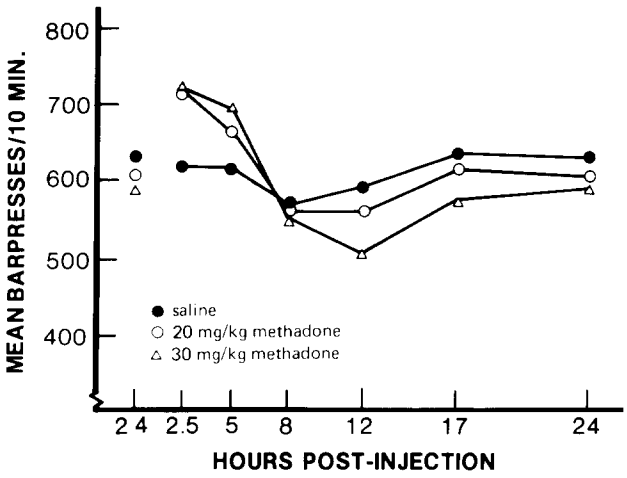

FIG. 2. Self-stimulation rates for the Drug Group following oral administration of saline, 20 mg/kg or 30 mg/kg of methadone (data collapsed over days). The means shown for 24 hours post-administration on either side of the graph represent the same data.

Response rates increased 2.5 and 5 hours post-administration, while rates decreased at several of the later testing times. In order to clarify the Group×Time interaction, *t*-tests were performed (data collapsed over days) to determine which of the testing times for the Drug Group were significantly different from the Saline Group. For both doses of methadone, the SS rates for the Drug Group were significantly higher ($p<0.05$) than those of the Saline Group at 2.5 hours post-administration. For the ME-30 treatment, the SS rates were significantly lower ($p<0.05$) than the Saline Group at 12 and 17 hours post-administration.

A three within (pre-post methadone Treatment, Days, and Time) ANOVA was performed to compare the effects of 15 mg/kg of morphine on SS in Experiments 1 and 2. No significant differences were obtained, indicating that the experience with methadone did not alter the animals' responsiveness to morphine.

Histology was performed on all animals which completed Experiment 2 and also on the five animals categorized as non-facilitators in Experiment 1. The electrode tips were located in the medial forebrain bundle or Forel's Field H_2. Exceptions were one electrode which was located in the substantia nigra, zona compacta, and two which were located in the substantia nigra, zona reticulata. The rostral-caudal levels were between A3750μ and A2420μ [11]. Differences in SS rate in response to opiates (between animals) were not observed to depend upon the locus of the electrode tip.

DISCUSSION

Methadone produced a facilitation of SS at 2.5 hours post-administration with the 20 mg/kg and the 30 mg/kg doses. The 20 mg/kg dose of orally administered methadone was much less effective than the equally potent [2,18] 10

mg/kg parenterally administered dose reported previously by this laboratory [25]. Also, with the doses examined here, the facilitation of SS with oral methadone was less than that obtained with parenteral morphine (see Fig. 1 and 2).

The existing literature on opiates and SS contains reports of suppression followed by facilitation of SS or simply a suppression with no facilitation [1,24]. These reports have generally attributed the suppression as being due to non-specific effects of opiates such as catatonia [28], which occur relatively early post-injection and show tolerance with repeated administrations. Since the lower response rates seen with the 30 mg/kg dose of methadone at 12 and 17 hours post-administration followed the facilitation and did not tolerate, another explanation may be required. The lower response rates may reflect a decrease in rewardability of the stimulation at these times or may simply be due to a mild withdrawal effect occurring between intubations. If these lower response rates represent a withdrawal effect, it is unclear why the rates do not progressively decrease up to the time of the next intubation.

GENERAL DISCUSSION

Facilitation of SS has been demonstrated following administration of parenteral morphine or oral methadone, which may be interpreted as being due to the reinforcing properties of these drugs. The greater facilitation of SS by parenteral morphine as compared to oral methadone may be explained in part by differences in pharmacokinetics associated with those routes of exposure. For example, oral administration of an opiate typically results in a delayed and lower peak effect [2]. That the facilitation with oral methadone is less than that previously observed with parenteral methadone [25] is consistent with the literature on human subjects demonstrating that the oral route is less effective in producing euphoria than is the subcutaneous route. These factors may contribute to the controversy in clinical reports concerning the possible euphoric properties of oral methadone [13], since delayed mild feelings of euphoria or well being may not readily be attributed to the drug.

The mild reinforcing properties of oral methadone may contribute to its relatively high acceptance rate and low dropout rate of addicts in maintenance programs. The reduction of intake of illicit narcotics by addicts in methadone maintenance programs is primarily due to the drug's ability to produce cross-tolerance to other narcotics while preventing withdrawal effects. However, since oral methadone itself produces some euphoria, the addict may have a reduced desire to supplement this with intake of other narcotics. Thus, in evaluating methadone maintenance and other treatment programs, it is important to consider the role of the euphoric properties of the pharmacological agent used.

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