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Chronic opioid exposure produces increased heroin self-administration in rats

John R. Walker[a], Scott A. Chen[a], Heather Moffitt[a], Charles E. Inturrisi[b], George F. Koob[a,*]

[a]*Department of Neuropharmacology, The Scripps Research Institute, CVN-7, 10550 North Torrey Pines Road, La Jolla, CA 92037, USA*
[b]*Department of Pharmacology, Cornell University Medical College, New York, NY, USA*

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Abstract

The purpose of this study was to determine the significance of chronic opioid exposure on the level of heroin self-administration in the rat. Rats were divided into morphine (M, subcutaneous morphine pellets) and placebo (P, subcutaneous placebo pellets) groups and self-administered several different doses of heroin during daily limited access 1-h sessions and prolonged access 8-h sessions. No effects on heroin self-administration occurred when the rats were implanted with morphine pellets and allowed to self-administer heroin in a limited access paradigm (1-h group). However, rats with morphine pellet implantation showed a rapid escalation (Days 0–3 post-pellet) in heroin self-administration in the more prolonged access group (8 h group) compared to placebo-pelleted animals also with 8-h access. Ultimately, placebo-pelleted 8-h exposed animals showed an escalation in heroin self-administration but this effect was delayed until Days 16–18 post-pellet. These results suggest that passive administration of morphine sufficient to produce and maintain dependence facilitates escalation in heroin intake.

Keywords: Heroin; Self-administration; Dependence; Morphine; Opioid; Rats

1. Introduction

Clinical evidence suggests that uncontrolled or excessive drug use is a key component of addiction to many drugs of abuse (Edwards, 1986; Marlatt and Baer, 1988). Dependence, defined as the manifestation of withdrawal symptoms after cessation of chronic drug intake, is believed to contribute to addiction (Jaffe, 1990). Understanding the relationship between dependence and uncontrolled drug use is of great importance to understanding and treating addiction (Marlatt and Baer, 1988).

It has been hypothesized that one factor that leads to excessive drug intake in dependent subjects is the motivation to alleviate withdrawal (Koob et al., 1989; Solomon and Corbit, 1974). Subcutaneous implantation of morphine pellets is a commonly used method to induce and maintain opioid dependence in rodents (Gold et al., 1994; Yoburn et al., 1985). If withdrawal motivates excessive drug intake, rats with subcutaneous morphine pellets implanted (morphine maintenance) would not be experiencing withdrawal and therefore would not increase opioid intake. Thus, withdrawal experienced after removal of morphine pellets might lead to drug intake above predependence values (Koob et al., 1989; Solomon and Corbit, 1974). Previous studies have shown that prolonged access to opiates via self-administration also leads to excessive drug taking (Ahmed et al., 2000; Sim-Selley et al., 2000). The purpose of this study was to determine whether concurrent morphine pellet implantation or deprivation from chronic opioid exposure leads to excessive heroin intake in the rat and to explore how this passive exposure compares to the escalation in intake observed in animals with prolonged access to the drug via self-administration.

2. Materials and methods

2.1. Subjects and surgical techniques

The subjects were 38 male Wistar rats, 180–200 g at the start of the experiments, housed in groups of two per cage, and maintained on a 12-h light/dark cycle (lights off at 6:00 p.m.). All self-administration sessions were performed dur-

* Corresponding author. Tel.: +1-858-784-7062; fax: +1-858-784-7405.
E-mail address: gkoob@scripps.edu (G.F. Koob).

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ing the rats' active (dark) phase. All rats were trained to lever-press for food on a fixed ratio 1 (FR 1) time-out (TO) 20-s schedule before implantation of jugular catheters. Catheters were implanted in all rats as described previously (Ahmed and Koob, 1997), and rats were allowed to recover for 7 days before the start of heroin self-administration sessions. Each day during recovery, rats were given intravenous injections of the antibiotic Timentin (SmithKline Beecham Pharmaceuticals, Philadelphia, PA), 0.1 ml of a 100-mg/ml solution. Each day throughout the experiment, catheters were flushed with 0.1 ml heparinized saline (3 U heparin) before and after the self-administration sessions. All experiments were performed in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals and were approved by the Institutional Animal Care and Use Committee of The Scripps Research Institute.

2.2. Heroin self-administration in dependent rats (1-h sessions)

Eighteen rats were allowed to self-administer heroin (10 μg/infusion) for 1 h/day for 9 days. Subsequently, they were tested with various doses of heroin presented in a Latin square design. Each rat received three sessions each at 5, 10, 20, and 40 μg heroin/infusion. Two groups with equivalent baseline self-administration rates were subsequently formed. One group (P rats) was implanted with placebo pellets, and the other group (M rats) was implanted with morphine pellets under light halothane anesthesia (1.5%). All pellets were standard 75-mg pellets (National Institute on Drug Abuse, Bethesda, MD), which were wrapped in nylon to ease subsequent removal. Self-administration sessions were continued with various doses of heroin presented in a Latin square design. Rats received three sessions each of 5, 10, 20, and 40 μg heroin/infusion (Days 1–15 post-pellet). The number of infusions per session for each dose was considered stable when the mean varied by less than ±10% over three sessions. All sessions were 1 h in duration to minimize the possibility of producing dependence in the P rats (Carrera et al., 1999). It has been shown previously that two subcutaneous morphine pellets release a stable level of morphine for at least 12 days (Gold et al., 1994). Therefore, old pellets were removed and new ones simultaneously implanted every 12–14 days during the course of the experiment. Rats were allowed one recovery day after each pellet replacement. One rat from the placebo group was eliminated from the experiment due to a blocked catheter.

2.3. Heroin self-administration in dependent rats (8-h sessions)

Another group of 25 rats was trained to self-administer heroin at a dose of 20 μg/infusion on an FR 1 TO 20-s schedule. Daily sessions were 2 h in duration, and the number of infusions during the session was recorded. The baseline number of infusions per session was considered stable when the mean varied by less than ±10% over three sessions. The mean ± S.E.M. number of infusions for the 2-h sessions was 12.5 ± 0.9. Rats were divided into three groups (placebo 2 h: 10 rats; placebo 8 h: 5 rats; morphine 8 h: 10 rats) with equivalent baseline self-administration rates. Rats in both placebo groups (P2 and P8 rats) were implanted with two subcutaneous placebo pellets as described above (Experimental Day 4). Rats in the morphine group were implanted with two subcutaneous morphine pellets (M8 rats) (Fig. 1). Three days after implantation (Experimental Days 4–6), heroin self-administration was continued for daily 8- (M8 rats and P8 rats) or 2-h sessions (P2 rats). P2 rats were limited to 2-h sessions to minimize the possibility of escalation in heroin intake developing over the course of the experiment. The rationale for the 2-h sessions in this experiment was that this was the standard session length for most experiments in this laboratory where no escalation occurs, and it was assumed that this session duration did not produce a dependent state.

2.4. Induction of repeated withdrawal and heroin dose-response

The goal of this experiment was to allow rats to self-administer heroin as withdrawal progressed and to avoid excessive withdrawal that might interfere with lever-pressing performance. Seven days after pellet implantation and after three self-administration sessions (Experimental Days 4–6) with the implanted pellets, pellets were removed (Fig. 1). After 3 h of recovery, P2 rats and M8 and P8 rats were continued on 2- and 8-h heroin self-administration sessions, respectively. All rats self-administered 20 μg heroin for three consecutive days (Experimental Days 7–9; Days 1–3 post-pellet) after the pellets were removed. Then, heroin doses (10, 20, 40, and 80 μg/infusion) were given in a Latin square design for three consecutive days at each dose (Experimental Days 10–24; Days 4–18 post-pellet). After rats had received all of the above doses, all rats self-administered a 160-μg/infusion dose of heroin for three consecutive days. M8 rats were given longer sessions to ensure that the onset of withdrawal coincided with heroin availability. P8 rats were given a long session to control for the amount of heroin self-administered by the M8 rats independent of morphine pellets and injections. To maintain high opioid blood levels, M rats were given 0.75 mg/kg morphine (whereas P2 and P8 rats received saline) in 1 ml/kg through the catheters immediately after the heroin self-administration session and again 6 h later (during the inactive light phase). The next heroin self-administration session began 6 h after the second injection. On rest days, rats were given 1.5 mg/kg morphine (M) or saline (P) through their catheters every 12 h (i.e., twice the daily supplemental dosage given in one dose). These conditions for supplemental morphine infusions were determined from analysis of blood morphine levels in rats after chronic pellet

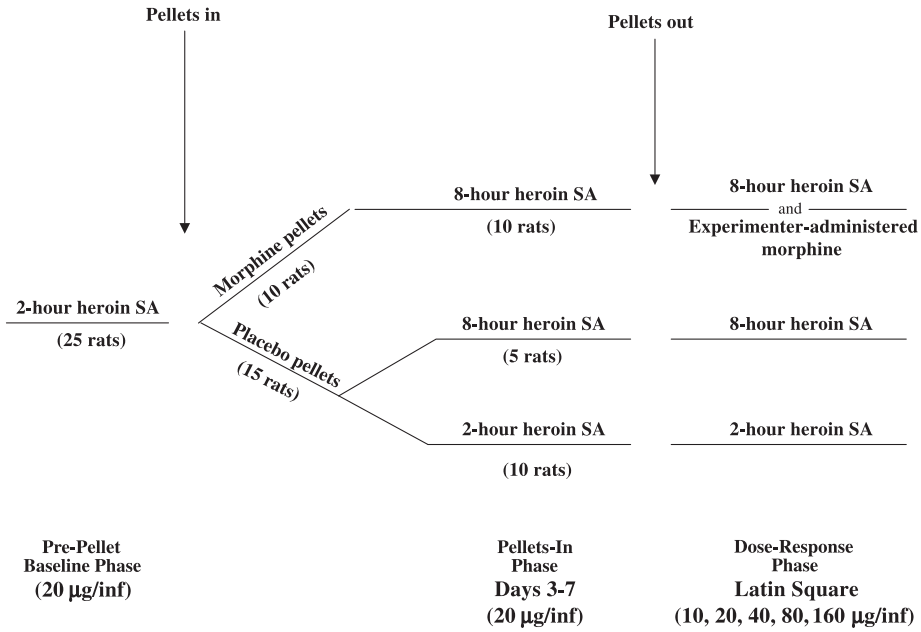

Fig. 1. Procedure to induce and maintain opioid dependence and repeated deprivation in rats self-administering heroin. Twenty rats self-administered a baseline dose of heroin (pre-pellet baseline phase) until responding was stable. Rats were divided into two groups: one group received morphine pellets and the other placebo pellets. Morphine-pelleted rats self-administered the baseline heroin dose for 8 h/day, while placebo-pelleted rats self-administered heroin for 2 h/day (“pellets in” phase). All pellets were removed, and rats previously pelleted with morphine self-administered five different heroin doses during 8-h sessions on separate days, 3 days for each dose. These rats were given twice-daily experimenter-administered morphine, 0.75 mg/kg iv. Previously placebo-pelleted rats self-administered heroin during 2-h sessions and were given experimenter-administered saline.

implantation and different doses and schedules of intravenous morphine administration (data not shown). Once pellets were implanted, and for the duration of the experiments, self-administration session were not performed every fourth day (i.e., no self-administration session) because progressive weight loss and diminishing self-administration rates were seen in the M rats. Weight loss was presumed to be related to the lack of available food during the longer self-administration sessions, and recovery of response rate and body weight coincided. Sessions were continued until the average number of infusions per hour was ±10% of the past three sessions, which took approximately 1 week.

2.5. Data analysis

The amount of heroin self-administered while pellets were implanted (number of infusions for a defined time period) was compared by two-way analysis of variance (ANOVA) with one between-subjects factor (experimental groups: placebo or morphine pelleted) and one within-subjects factor (pre-pellet and with pellet). The number of heroin infusions and amounts of heroin self-administered post-pellet removal were compared by two-way ANOVA with one between-subjects factor (experimental groups: placebo 2 h [P2 group], placebo 8 h [P8 group], or morphine-pelleted [M8 group]) and with repeated measures on the second factor (dose in mg/kg/injection or μg heroin/injection). Post hoc comparisons were carried out using tests for simple main effects. Mean body weights of animals were compared between groups during the Latin square dose-response phase of each study, and differences between groups were compared with a one-way ANOVA followed by a Newman–Keuls post hoc test.

3. Results

3.1. Heroin self-administration: concurrent pellet implantation

M or P rats showed no difference in the amounts of four different doses of heroin consumed before pellets were implanted. Specifically, morphine-pelleted rats self-administered (mean ± S.E.M.) 254.4 ± 39.4, 280.7 ± 46.5, 336.8 ± 37.2, and 473.5 ± 61.2 μg/kg of 5, 10, 20, and 40 μg doses of heroin before pellets were implanted. While pellets were in, the amount of heroin consumed of the same doses was not significantly different (mean ± S.E.M.): 200.5 ± 31.4, 234.4 ± 28.1, 311.9 ± 24.4, and 412.4 ± 28.3 [$F(3,48) = 0.170$, ns]. Finally, no differences were seen between the morphine- and placebo-pelleted rats while pellets were implanted [$F(3,45) = 0.275$, ns] (Fig. 2).

3.2. Heroin self-administration: effect of deprivation state

Heroin self-administration in a separate group of rats undergoing opioid deprivation (M rats) was compared to that of nondeprived rats (P2 and P8 rats). Following a significant

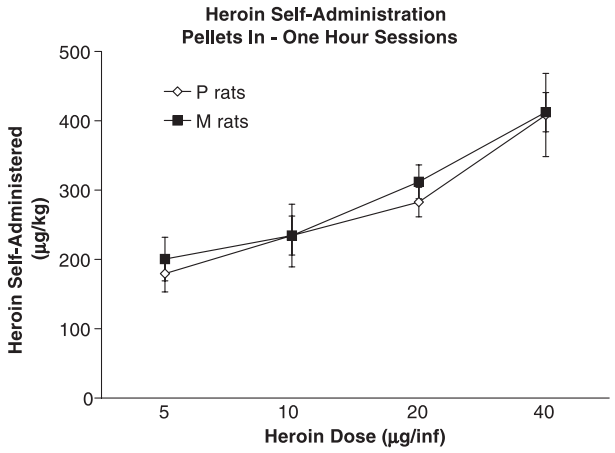

Fig. 2. Dose-response of heroin self-administration comparing morphine (M rats) vs. placebo (P rats) pelleted rats while pellets were implanted. Data are expressed as micrograms of heroin self-administered per kilogram during the 1-h session. No between-group differences were seen [$F(3,45) = 0.096$, ns].

one-way ANOVA [$F(2,22) = 12.158$, $P < .001$], the body weight of M8 rats was on average 87% that of P2 rats on the last day of pellet implantation (M8 rats: 393.4 ± 5.7 g; P2 rats: 450.2 ± 11.7 g, $P < .001$, Newman–Keuls post hoc test). The body weight of M8 rats was not significantly different from that of P8 rats (378.2 ± 17.6). Data therefore were expressed as micrograms per kilogram of heroin consumed over the first 2 h of the sessions for both groups. A significant increase was seen in the amount of heroin self-administered while morphine pellets were still implanted (M rats) while no effect was seen in P2 or P8 rats (Fig. 3). A significant Group

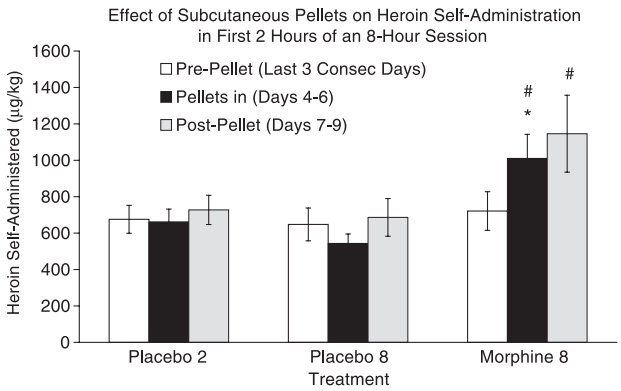

Fig. 3. Effect of subcutaneous pellet implantation on heroin self-administration. Rats implanted with morphine pellets self-administered more heroin (20 µg/infusion) than rats implanted with placebo pellets. Amount of heroin consumed is expressed as micrograms per kilogram body weight over the first 2 h of the self-administration session. A significant Group × Condition (pre-pellet vs. pellets in vs. post-pellet) interaction occurred [$F(4,44) = 4.29$, $P < .01$] with a significant difference between the placebo and morphine pellet groups with the pellets implanted [$F(1,18) = 6.02$, $P < .05$]. Within the morphine group, pellet implantation significantly increased the number of heroin self-infusions [$F(2,18) = 10.22$, $P < .01$]. Significant increases occurred with pellets implanted ($P < .05$, Bonferroni/Dunn test) and after pellets were removed ($P < .05$, Bonferroni/Dunn test). * $P < .05$ vs. placebo "in"; $^{\#}P < .05$ vs. morphine "pre".

(P2 vs. P8 vs. M8) × Condition (pre-pellet vs. with pellet vs. post-pellet) interaction was observed [$F(4,44) = 4.29$, $P < .01$], with a significant increase ("pellets in" condition) in the morphine-pelleted group compared to the placebo group [$F(1,18) = 6.02$, $P < .05$]. In the M8 rats, a significant increase in the number of heroin self-infusions compared to the pre-pellet condition was seen [$F(2,18) = 10.22$, $P < .01$] when the pellets were still implanted ($P < .05$, Bonferroni/Dunn test) and after they were removed ($P < .05$, Bonferroni/Dunn test).

After removal of pellets, M8 rats self-administered significantly more of various heroin doses than P2 rats. A significant Dose × Treatment interaction was observed [$F(8,88) = 11.16$, $P < .001$]. A significant simple effect for treatment was found at three doses: 40 µg/infusion [$F(2,22) = 6.34$, $P < .01$], 80 µg/injection [$F(2,22) = 4.83$, $P < .05$], and 160 µg/injection [$F(2,22) = 11.61$, $P < .001$], indicating differences between the three treatment groups at each dose (see Fig. 4). Subsequent simple comparisons revealed that, compared to P2 controls, M8 rats self-administered significantly more of the 40 ($P < .01$, Bonferroni/Dunn test), 80 ($P < .01$, Bonferroni/Dunn test), and 160 µg/injection ($P < .001$, Bonferroni/Dunn test) doses, while the P8 rats self-administered significantly more of the 160 µg/injection ($P < .001$, Bonferroni/Dunn test) dose. A significant two-way mixed ANOVA ([$F(8,88) = 19.51$, $P < .0001$) revealed similar increases in heroin self-administration in the first half-hour for M8 and P8 rats over P2 rats, and in the amount of heroin consumed per hour [$F(8,88) = 9.13$, $P < .0001$]. The total amounts of opiate administered to and consumed by the M and P rats are shown in Table 1.

Rats were checked for classical somatic withdrawal signs throughout all experiments, and no signs were observed.

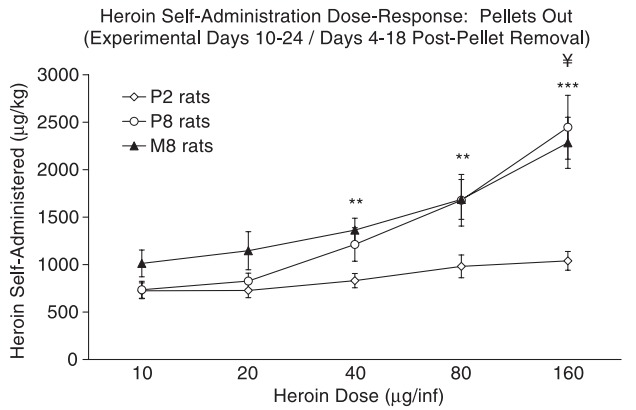

Fig. 4. Self-administration of different heroin doses, expressed in micrograms heroin consumed per kilogram body weight during the first 2 h of each session, in previously placebo-pelleted (P2 or P8) or previously morphine-pelleted (M) rats. The P2 group had a total of a 1-h session. The P8 and M groups had a total of an 8-h session. Three consecutive sessions were performed for each dose of heroin. The Latin square dose-response design was used for the first four doses (Days 4–15 post-pellet), after which all rats then received the highest dose of 160 µg/infusion (Days 16–18 post-pellet). ** $P < .01$, *** $P < .001$, difference between M8 and P2; $^{¥}P < .001$, difference between P8 and P2.

Table 1
Total opioid exposure of M and P rats

	P2 rats	P8 rats	M8 rats
Amount of heroin self-administered (mg/kg)[a]	12.9 ± 1.2	80.98 ± 8.0	92.5 ± 11.9
Amount of morphine administered by experimenter (mg/kg)[+]	0	0	30

[a] Rats self-administered heroin during 8- (M8 and P8 rats) or 2-h (P2 rats) sessions during the entire experimental period. Results are expressed as mean ± S.E.M.

[+] Rats received intravenous infusions of 0.75 mg/kg morphine (M rats) or saline (P8 and P2 rats) twice daily for 20 days. Rats also received 2 × 75-mg sc morphine (M rats) or placebo (P8 and P2 rats) pellets for 7 days.

Some gnawing behavior was detected periodically in the majority of rats and appeared unrelated to the amount of heroin self-administered or to the treatment group.

4. Discussion

Subcutaneous morphine pellets had no effect on the amounts of heroin self-administered while the pellets were still implanted if access was limited (1 h/day). In contrast, morphine-pelleted rats showed a more rapid increase in heroin intake with prolonged sessions compared to placebo-pelleted rats also in prolonged sessions. These results suggest that the passive exposure of morphine accelerates escalated intake in animals only when paired with prolonged sessions.

The lack of an effect of chronic morphine pellet implantation on heroin self-administration in a limited access situation is consistent with previous studies. Methadone and morphine maintenance have been shown previously in dogs to have no lasting effect on morphine self-administration after an initial adjustment period (Jones and Prada, 1977). Our results extend these studies by adding multiple self-administered opioid doses.

However, with extended access to heroin via self-administration, rats with chronic morphine pellets dramatically increased their heroin intake. M8 rats self-administered more of a single dose of heroin than P2 or P8 rats, and more than their pre-pelleted condition when pellets were implanted. Under these conditions, rats likely were not experiencing withdrawal at any time during their self-administration sessions, and withdrawal could not have contributed to the observed increase in intake.

The factor that resulted in increased heroin self-administration in morphine-pelleted rats with prolonged access to heroin self-administration could have been the prolonged self-administration sessions and not previous implantation of subcutaneous morphine pellets (compare Figs. 2–4). We have shown previously that prolonged heroin self-administration sessions *alone* result in increased heroin self-administration (Ahmed et al., 2000). A more likely explanation is that there is an interaction between morphine pellet history and prolonged access. In the previous study, escalation required 7–11 days (see Figs. 1 and 3 of Ahmed et al., 2000). Indeed, in the present study, placebo-pelleted rats that experienced twenty-one 8-h self-administration sessions ultimately escalated their heroin intake as was demonstrated in Fig. 4 and by the observation that their baseline ultimately shifted to an escalated state.

However, rats in an opioid-dependent state (Experimental Days 7–9; Days 1–3 post-pellet) initially self-administered more heroin than rats with a more limited history of opioid administration (P2 or P8 rats). There are several reasons for this phenomenon. One possible explanation for the increase in heroin self-administration in opioid-deprived rats (compared to their pre-pelleted condition and to P rats) is that they experienced spontaneous withdrawal and self-administered heroin to attempt to alleviate withdrawal distress. Rats were examined for physical withdrawal symptoms at the same time they started their daily sessions, and no differences were seen between dependent and non-dependent rats (data not shown). Nevertheless, previous work has shown that the signs of opioid withdrawal that model negative affective states (e.g., increased brain stimulation reward thresholds, place aversion, and so on) are manifest before physical signs become apparent (Schulteis et al., 1994).

Thus, the present results show increased heroin intake in animals with the pellets in and with the pellets out if the animals have prolonged access to the drug (8-h access) but no increased heroin intake in limited access situations (1- to 2-h access) with or without pellets. One explanation for increased heroin intake in rats with 8-h access could be tolerance to heroin's reinforcing effects. M rats consumed approximately seven times more heroin, and P8 rats consumed approximately six times more heroin than P2 rats over the course of the study, presumably sufficient to induce tolerance (see Table 1). There is some evidence that tolerance can develop to the reinforcing effects of opioids (Shippenberg et al., 1988), but a lack of tolerance and also sensitization have been reported using place conditioning (Contarino et al., 1997; Lett, 1989) and brain stimulation reward (Kornetsky and Duvauchelle, 1994). The difference in these results may depend in part on the schedule of opioid administration (Contarino et al., 1997; Lett, 1989; Shippenberg et al., 1988). Tolerance to a drug's effects is reflected in a parallel shift to the right of the dose–effect curve for that drug (change in sensitivity), with no change in the height of the curve (Levine, 1990). Conversely, sensitization could explain a parallel shift to the left in the dose–effect curve. The present results reflect more of an upward shift of the dose–effect curve as previously described (Ahmed and Koob, 1998, 1999) and therefore may represent an increase in the reinforcing efficacy of heroin during abstinence (Fig. 4).

The behavioral data presented here suggest that prolonged exposure to opiates leads to escalated heroin self-administration when the prolonged exposure is due to the session length and not the morphine pellets alone. Morphine pellet exposure simply speeds up the escalation process

when paired with prolonged session length. The neurobiological basis for this increased drug intake may be key to our understanding of the changes in the brain that lead to opiate dependence.

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References

Ahmed SH, Koob GF. Cocaine- but not food-seeking behavior is reinstated by stress after extinction. Psychopharmacology 1997;132:289–95.

Ahmed SH, Koob GF. Transition from moderate to excessive drug intake: change in hedonic set point. Science 1998;282:298–300.

Ahmed SH, Koob GF. Long-lasting increase in the set point for cocaine self-administration after escalation in rats. Psychopharmacology 1999; 146:303–12.

Ahmed SH, Walker JR, Koob GF. Persistent increase in the motivation to take heroin in rats with a history of drug escalation. Neuropsychopharmacology 2000;22:413–21.

Carrera MR, Schulteis G, Koob GF. Heroin self-administration in dependent Wistar rats: increased sensitivity to naloxone. Psychopharmacology 1999;144:111–20.

Contarino A, Zanotti A, Drago F, Natolino F, Lipartiti M, Giusti P. Conditioned place preference: no tolerance to the rewarding properties of morphine. Naunyn Schmiedebergs Arch Pharmacol 1997;355: 589–94.

Edwards G. The alcohol dependence syndrome: a concept as stimulus to enquiry. Br J Addict 1986;81:171–83.

Gold LH, Stinus L, Inturrisi CE, Koob GF. Prolonged tolerance, dependence and abstinence following subcutaneous morphine pellet implantation in the rat. Eur J Pharmacol 1994;253:45–51.

Jaffe JH. Drug addiction and drug abuse. In: Gilman AG, Rall TW, Nies AS, Taylor P, editors. Goodman and Gilman's the pharmacological basis of therapeutics. 8th ed. New York: Pergamon; 1990. p. 522–73.

Jones BE, Prada JA. Effects of methadone and morphine maintenance on drug-seeking behavior in the dog. Psychopharmacology 1977;54: 109–12.

Koob GF, Stinus L, Le Moal M, Bloom FE. Opponent process theory of motivation: neurobiological evidence from studies of opiate dependence. Neurosci Biobehav Rev 1989;13:135–40.

Kornetsky C, Duvauchelle C. Dopamine, a common substrate for the rewarding effects of brain stimulation reward, cocaine, and morphine. NIDA Res Monogr 1994;145:19–39.

Lett BT. Repeated exposures intensify rather than diminish the rewarding effects of amphetamine, morphine, and cocaine. Psychopharmacology 1989;98:357–62.

Levine RR. Pharmacology: drug actions and reactions. 4th ed. Boston: Little, Brown; 1990.

Marlatt GA, Baer JS. Addictive behaviors: etiology and treatment. Annu Rev Psychol 1988;39:223–52.

Schulteis G, Markou A, Gold LH, Stinus L, Koob GF. Relative sensitivity to naloxone of multiple indices of opiate withdrawal: a quantitative dose-response analysis. J Pharmacol Exp Ther 1994;271:1391–8.

Shippenberg TS, Emmett-Oglesby MW, Ayesta FJ, Herz A. Tolerance and selective cross-tolerance to the motivational effects of opioids. Psychopharmacology 1988;96:110–5.

Sim-Selley LJ, Selley DE, Vogt LJ, Childers SR, Martin TJ. Self-administration desensitizes μ opioid receptor-activated G-proteins in specific regions of rat brain. J Neurosci 2000;20:4555–62.

Solomon R, Corbit J. An opponent-process theory of motivation: I. Temporal dynamics of affect. Psychol Rev 1974;81:119–45.

Yoburn BC, Chen J, Huang T, Inturrisi CE. Pharmacokinetics and pharmacodynamics of subcutaneous morphine pellets in the rat. J Pharmacol Exp Ther 1985;235:282–6.