

RAPID COMMUNICATION

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Sensitization to the motor effects of contingent infusions of heroin but not of κ agonist RU 51599

Received: 27 December 1997 / Final version: 8 May 1998

Abstract It has been postulated that behavioral sensitization could reflect drug-induced changes that are central to the development of drug abuse; however, it is still unknown whether behavioral sensitization can arise during a “voluntary” and “self-controlled” consumption of drugs and consequently play a role in drug abuse. For this reason, we studied the possible sensitization of motor activity during ten consecutive intravenous self-administration (SA) sessions of one of the most largely abused opiates the μ agonist heroin [30 μ g/infusion (inf)]. We also studied in similar conditions the new κ agonist RU 51599 (6.5, 20 and 100 μ g/inf). Heroin and RU 51599 were compared because both drugs are self-administered by rodents, but the reinforcing properties of RU 51599 are very weak compared to those of heroin. At low ratio requirement rats developed SA of both heroin and RU 51599; however, a progressive increase in drug-induced locomotor activity over subsequent sessions was observed only for heroin but not for RU 51599. Sensitization of the motor effects of heroin developed over a period of time during which the intake of the drug was constant. In conclusion, sensitization can develop during the voluntary consumption of an addictive drug such as heroin. These results are in line with the hypothesis that sensitization could play a role in the etiology of drug abuse.

Key words Behavioral sensitization · Self-administration · Locomotion · Heroin · κ agonists · RU 51599

Introduction

The phenomenon of “behavioral sensitization” refers to a progressive increase in the response to psychoactive drugs following their repeated administration. Behavioral sensitization of the psychomotor effects of drugs is classically observed after administration of psychostimulants, such as cocaine and amphetamine, as well as after opiates, such as morphine (for review see Robinson and Becker 1986; Kalivas and Stewart 1991). Recently, it has been postulated that this phenomenon could reflect drug-induced changes playing an important role in the development of drug abuse (Piazza et al. 1989; Robinson and Berridge 1993). Indeed, both cocaine (Horger et al. 1990) and amphetamine (Piazza et al. 1989, 1990) self-administration (SA) are more readily developed if subjects are repeatedly pre-exposed to the self-administered drug. Furthermore, repeated administration of psychostimulants or opiates also increases the rewarding properties of these drugs as measured by the conditioned place preference paradigm (Lett et al. 1989).

Most papers studying the phenomenon of behavioral sensitization to the psychomotor stimulant effects of drugs have considered changes in behavioral activity after non-contingent drug injections. This leaves it debatable whether behavioral sensitization can arise from a “voluntary” and “self-controlled” consumption of drugs. This is a relevant question, since there is evidence that the neurochemical and behavioral effects of drugs can be different if they are contingently or non-contingently administered (Wilson et al. 1994; Dworkin et al. 1995; Hemby et al. 1995).

Recently, two papers have partially addressed this question reporting the development of sensitization to the locomotor effects of a non-contingent challenge injection of cocaine in subjects that have been self-administering this psychostimulant (Hooks et al. 1994; Phillips and Di Ciano 1996). However, it is still unknown if the behavioral effects of voluntary injections

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of psychostimulants or opiates sensitize *during* their self-controlled consumption.

For this reason, we studied the possible sensitization of motor activity during voluntary consumption of the most abused opiate, the μ agonist heroin, by motoring locomotor activity during intravenous SA of this drug. In similar experimental conditions, we also studied the new κ agonist RU 51599 (Gueniau and Oberlander 1997; Hamon et al. 1995). Heroin and RU 51599 were compared because, although we have recently shown that both drugs are self-administered by rodents (Marinelli et al. 1996), their pharmacological profiles differ profoundly. First, compared to heroin, RU 51599 has weak reinforcing properties, since animals only self-administer it at very low ratio requirements, and, similarly to other κ agonists (for review see Woods and Winger 1987; Ramsey and Van Ree 1992), it induces place aversion instead of place preference (Marinelli et al. 1996). Second, RU 51599 decreases dopamine release in the nucleus accumbens, whereas heroin increases it (Marinelli et al. 1996). In conclusion, RU 51599 does not have the profile of an addictive drug. Consequently, the comparison of RU 51599 and heroin could provide some insight on the possible link between the appearance of sensitization and the addictive properties of a pharmacological compound.

Materials and methods

Subjects

Male Sprague–Dawley rats (Iffa Credo; Lyon, France) weighing 300–320 g were used. A 12:12 dark/light cycle (lights off 12 p.m.) was maintained in the animal room, and temperature (22°C) and humidity (66%) were kept constant. Animals were housed individually and allowed at least 1 week to acclimatize to the animal room before starting any manipulation. During this time, animals had ad libitum access to food and water. Following this acclimatization period, animals were progressively brought to 90% of their body weight by decreasing their daily ration of food. The daily ration of food was given at 4 p.m. and animals were food-restricted for at least 12 days before the start of any other experimental manipulation, during which food restriction was maintained.

The procedures described here comply with ethical principles and guidelines for care and use of laboratory animals adopted by the European Community, law 86/609/CEE.

Intravenous self-administration

Animals ($n = 5–6$ per group) were submitted to surgery under ether anaesthesia. A silastic catheter (20 μ l dead volume) was inserted in the right auricle through the external jugular vein, passed under the skin and fixed in the mid-scapular region. After 1 week of recovery, independent groups of animals were tested for SA of heroin (heroin HCl, Francopia, Gentilly, France) 30 μ g/inf, or RU 51599 (kindly donated by Roussel Uclaf, Romanville, France) 6.5 μ g, 20 μ g or 60 μ g/inf. These doses were chosen because they readily induce SA of the two compounds. Both drugs were dissolved in sterile 0.9% NaCl solution and each infusion had a volume of 20 μ l. Rats were submitted to daily SA sessions during the dark period (1 p.m.) for a period of 1 h. Before the start of each session, the

catheter was filled with 20 μ l of the heroin or RU 51599 solution, according to the experimental group, and its external end was connected to a pump-driven syringe. The SA cage (35 \times 33 cm floor area, 50 cm high) was equipped with two holes located 6 cm above the floor and placed symmetrically in the center of two opposite walls. The introduction of the animal's nose (nose-poke) in one of the holes (defined as active), switched on the injection pump which delivered, according to the experimental group, 20 μ l of the heroin or RU 51599 solutions over a period of 2 s. Subsequent nose-pokes during the infusion period had no effects on the injection pump, but were recorded. Nose-pokes in the other hole (defined as inactive) had no effects at any time. The number of nose-pokes in both holes and the number of infusions were recorded throughout the experiments. At the end of each session, the catheter was filled with 100 μ l of a heparin-streptokinase solution (10 and 3000 IU, respectively) to prevent from clogging.

During the first 5 days of testing for SA, a fixed ratio (FR) of 1 was used, i.e. one infusion was delivered per nose poke. Following this acquisition period, the number of nose-pokes required to obtain one infusion was progressively increased from one to five: FR3 (3 days), FR5 (2 days). The comparison of the number of nose pokes between the active and inactive holes was used to determine if animals acquired the task (i.e. self-administered). An animal was considered to develop SA when the number of nose-pokes in the active hole was significantly higher than the number of nose pokes in the inactive one.

Locomotor activity

Locomotor activity was monitored throughout each SA session by means of two photocells located 2 cm above the floor and placed 10 cm from each side of the active and inactive holes of the SA cages. A motor activity count was registered each time the rat broke one of the two beams (total counts). The number of times the animal crossed the full distance between the photocells (15 cm), breaking the two beams in succession, was also recorded (crossings).

Statistical analyses

All data were subjected to an analysis of variance (ANOVA) for repeated measures. To attest SA, the number of nose pokes in the active and inactive hole was used as a first within factor (Hole effect, two levels) and the different SA sessions were used as a second within factor (Days effect, five levels in the acquisition phase, five levels for the remaining sessions and ten levels throughout the entire experiment). To verify if the animals maintained a stable behavior throughout the experiment, the number of infusions and the locomotor activity (both total counts and crossings) were also analyzed over the days. For brevity, only results relative to the number of self-injections and total activity counts have been graphed.

Results

Heroin self-administration

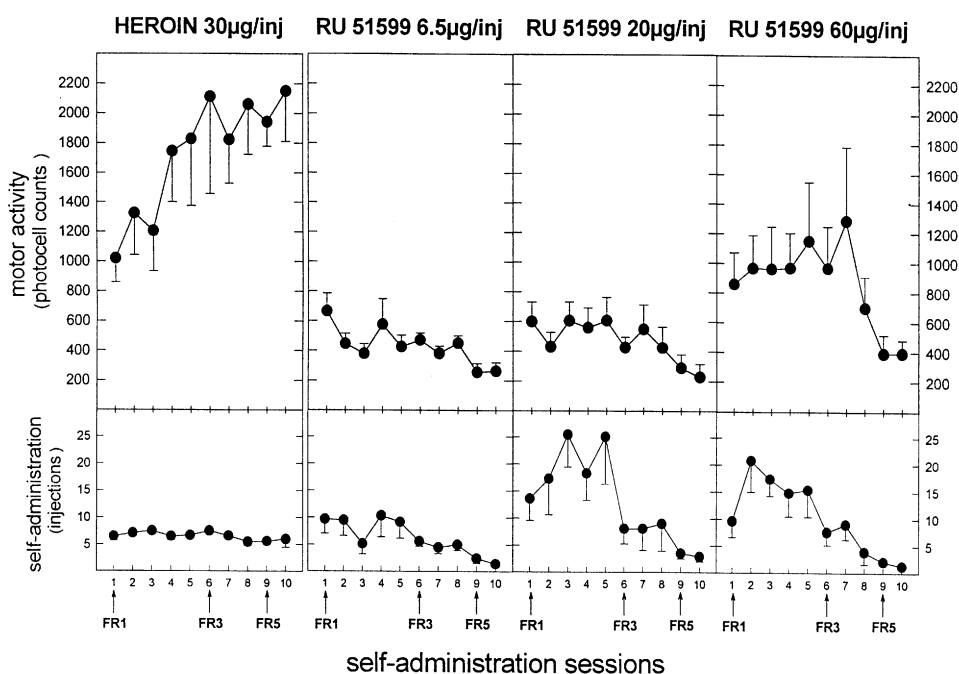
All animals developed SA and maintained this behavior throughout the experiment. Thus, the number of nose pokes in the active hole was significantly higher than the number of nose-pokes in the inactive one both in the acquisition phase (Hole effect, $F[1,5] = 41.75$, $P < 0.001$) and throughout the remaining five SA sessions (Hole effect, $F[1,5] = 35.98$, $P < 0.002$). The

increase in ratio induced a progressive increase in the number of nose-pokes in the active hole (from 9 ± 0.9 at FR1 to 26 ± 4.6 at FR5) (Days effect, $F[9,45] = 5.7$, $P < 0.001$), without significant changes in the number of responses in the inactive hole. Animals showed a constant intake of heroin throughout the entire experiment, regardless of the increase in ratio to obtain the reinforcement (Fig. 1a, bottom panel). Thus, the number of self-infusions did not vary either during the first 5 days of testing at FR1 (Days effect, $F[4,20] = 0.62$, $P < 0.65$) or over the entire SA sessions (Days effect, $F[9,45] = 0.92$, $P < 0.5$). Despite the constant intake of the drug, there was a progressive increase in drug locomotor effects as shown by the increase in both the total counts (Days effect, $F[9,45] = 3.01$, $P < 0.01$) and the crossings (Days effect, $F[9,45] = 2.25$, $P < 0.05$) over the 10 days of SA (Fig. 1a top panel). Such an increase in motor activity was already present during the first 5 days of SA (Days effect, $F[4,20] = 3.41$, $P < 0.03$ and $F[4,20] = 3.75$, $P < 0.02$ for the crossings and total counts, respectively) and no further increase was observed subsequently.

Fig. 1 Locomotor activity during heroin and RU 51599 self-administration. *Arrows* represent the start of a given ratio requirement (FR). Animals presented a constant intake of heroin (30 $\mu\text{g}/\text{inj}$) (*bottom*) throughout the 10 SA session; however, there was a progressive increase in locomotor activity during SA (*top*). This sensitization developed during the first 5 days of testing. SA of RU 51599 at 6.5, 20, and 60 $\mu\text{g}/\text{inj}$ was fairly constant during the first five sessions at FR1 and decreased when the ratio requirement was increased (*bottom*). Locomotor activity in these animals was constant during the first five sessions when the intake of the drug was constant, and decreased later on, in parallel with the decrease in drug intake

RU 51599 self-administration

As reported previously (Marinelli et al. 1996), animals developed SA at all doses of RU 51599 at a low schedule of reinforcement, but extinguished this behavior as the number of requests to obtain the drug increased. Thus, for all the doses studied, the number of nose pokes in the active hole was significantly higher than the number of nose-pokes in the inactive one only during the acquisition phase at FR1 (Hole effect, $F[1,5] = 10.76$, $P < 0.02$; $F[1,5] = 6.78$, $P < 0.02$ and $F[1,4] = 7.7$, $P < 0.05$ for the 6.5, 20 and 60 $\mu\text{g}/\text{inj}$ doses, respectively) but not during the remaining five sessions at FR3 and 5 (Hole effect, $F[1,5] = 4.1$, $P > 0.1$; $F[1,5] = 2.24$, $P > 0.2$ and $F[1,4] = 4.5$, $P > 0.1$ for the 6.5, 20 and 60 $\mu\text{g}/\text{inj}$ doses, respectively). Furthermore, as Fig. 1 shows, for all doses of RU 51599, animals only exhibited a constant intake of the κ agonist during the FR1 schedule, but decreased the intake throughout the SA sessions when the ratio to obtain the drug increased (Days effect, $F[9,45] = 2.38$, $P < 0.03$; $F[9,45] = 2.83$, $P < 0.01$ and $F[9,36] = 5.1$, $P < 0.001$ for the 6.5, 20 and 60 $\mu\text{g}/\text{inj}$ doses, respectively). When the intake of the drug was constant, during the first 5 days of testing, no change in locomotor activity was observed for any of the doses (Days effect, crossings: $F[4,20] = 1.5$, $P < 0.2$; $F[4,20] = 0.63$, $P < 0.78$ and $F[4,16] = 0.90$, $P < 0.48$; total counts: $F[4,20] = 1.48$, $P > 0.24$; $F[4,20] = 0.54$, $P > 0.71$ and $F[4,16] = 0.80$, $P > 0.54$ for the 6.5, 20 and 60 $\mu\text{g}/\text{inj}$ doses, respectively). Furthermore, throughout the 10 days of RU 51599 SA, the locomotor effects of this drug decreased or had a tendency to decrease (Days effect, crossings: $F[9,45] = 4.29$, $P < 0.001$; $F[9,45] = 1.94$, $P = 0.07$ and $F[9,36] = 2.7$, $P < 0.02$;



total counts: $F[9,45] = 2.45$, $P < 0.03$; $F[9,45] = 2.05$, $P = 0.055$ and $F[9,36]$, $P < 0.02$ for the 6.5, 20 and 60 $\mu\text{g}/\text{inf}$ doses, respectively). Such a decrease was parallel to the decrease in the number of self-injections.

Discussion

The results of this report show that the locomotor effects of intravenously self-infused heroin sensitize over time. Thus over 5 days of intravenous heroin SA, during which a stable intake of the drug was maintained, locomotor activity progressively increased over the sessions. Although previous results have shown that non-contingent repeated drug administration induces an increase in the psychomotor effects of opiates (Babbini and Davis 1972; Shuster et al. 1975; Joyce and Iversen 1979; Vezina et al. 1987), it is the first time, to our knowledge, that this effect is described during spontaneous drug intake. In contrast, though animals self-administered the κ agonist RU 51599, no sensitization of its motor effects was observed. As reported elsewhere (Marinelli et al. 1996), compared to heroin, RU 51599 showed weak reinforcing effects because its intake was not maintained when the ratio requirement was increased.

It has been postulated that the increase in drug response following repeated exposure, reflects drug-induced changes that are important for the establishment of addiction (Robinson and Berridge 1993). The present results are in line with this view, since self-administration of the largely abused opioid heroin induced a progressive sensitization of the motor effects of this drug. The absence of sensitization during self-administration of the κ agonist RU 51599 allows to speculate that there could be a specific link between sensitization and the addictive properties of a pharmacological compound. Thus, compared to heroin RU 51599 showed much weaker positive reinforcing effects.

An increase in the activity of the mesencephalic dopaminergic transmission, and especially of the dopaminergic projection to the nucleus accumbens, has been considered to be an important neural substrate of the addictive properties of drugs (for review, see Fibiger and Phillips 1988; Wise and Rompré 1989) and of drug-induced sensitization (Robinson and Berridge 1993). Our results are in line with these ideas. Thus, heroin, which showed strong reinforcing properties and induced sensitization, is known to increase dopamine release in the nucleus accumbens (Kalivas and Duffy 1988; Tanda et al. 1997). In contrast, RU 51599, which had weak reinforcing properties and did not induce sensitization, similarly to other κ agonists (Di Chiara and Imperato 1988a,b), has been shown to decrease dopamine release in the nucleus accumbens (Marinelli et al. 1996).

In conclusion, behavioral sensitization to the psychomotor activating properties of abused opiates

develops during a stable and voluntary self-intake. These results are in line with the idea that sensitization could reflect a drug-induced change playing an important role in the development of drug abuse.

Acknowledgements We would like to thank T.E. Robinson for helpful comments on an earlier version of this manuscript. This work was supported by INSERM, Université de Bordeaux II, Conseil Régional d'Aquitaine, Pôle Médicament Aquitaine.

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