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Intracerebroventricular Naltrexone Treatment Attenuates Acquisition of Intravenous Cocaine Self-Administration in Rats

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RAMSEY, N. F. AND J. M. VAN REE. *Intracerebroventricular naltrexone treatment attenuates acquisition of intravenous cocaine self-administration in rats.* PHARMACOL BIOCHEM BEHAV **40**(4) 807–810, 1991.—The influence of centrally administered naltrexone, an opiate antagonist, on acquisition of intravenous cocaine self-administration behaviour in rats was examined. On five consecutive days, three hours per day, they could self-administer a cocaine solution (30 μg per infusion) through an indwelling cannula. Treatment consisted of daily injections of naltrexone (2 or 5 μg) or placebo into the lateral ventricle 30 minutes before testing. Naltrexone treatment dose dependently attenuated the rate of cocaine self-infusion. Both self-infusion rate and rate of responding on the reinforcement lever in the group treated with 5 μg naltrexone differed from placebo, whereas rate of responding on a dummy lever did not. These findings a) support the notion that opioid systems play a role in cocaine reinforcement, and b) suggest that naltrexone exerts its effect on cocaine reinforcement through action in the central nervous system.

Cocaine Naltrexone Intracerebroventricular Self-administration Opioid systems

COCAINE is a psychomotor stimulant drug with reinforcing properties. Cocaine inhibits reuptake of serotonin (33), norepinephrine (16) and dopamine (17). The motor stimulating but also the reinforcing effects are attributed to cocaine's ability to increase extracellular dopamine (DA) levels in the mesocorticolimbic system (24,26). Activation of DA activity in this system is seen by some as the essential neurochemical process underlying the reinforcing effects of most if not all drugs of abuse (40). Other systems may also contribute to the rewarding effects of cocaine, such as serotonergic (7,23) and opioid systems (10, 11, 25). Opioid systems have been implicated in reinforcement of opiates (36), alcohol (1,12) and intracranial electrical self-stimulation (28,39), suggesting a role of endorphins in reinforcement processes per se (19,36).

Opioid involvement in cocaine reinforcement has been shown in a number of reports on cocaine self-administration behaviour. Carroll et al. (10) found that systemic NTX treatment caused an increase in self-administration rate of cocaine in trained rats. This was taken to reflect an attenuation of cocaine's rewarding effects, leading to a compensatory increase in intake. De Vry et al. (11) then examined the effect of NTX treatment on the acquisition of intravenous cocaine self-administration behaviour in rats, and found an attenuation. They tested the NTX effect with several unit doses of cocaine, and their results suggest that NTX caused a rightward shift in the dose-response curve. This supports the notion that opioid blockade results in decreased reinforcing effects of cocaine. Ramsey et al. (25) tested the postulated opioid involvement in cocaine reward further. They looked at the acquisition of intravenous self-administration of several cocaine unit doses after a period of chronic opioid blockade. In this study a leftward shift occurred in the cocaine dose-response curve, suggesting enhancement of cocaine reinforcement. They brought forward the possibility that opioid receptor upregulation may underly this effect of NTX pretreatment. Ettenberg et al. (13) failed to show an effect of NTX on cocaine self-administration. Their finding may, however, be explained by the fact that they used a comparably high cocaine unit dose (approximately 0.25 mg/infusion). In both De Vry and Ramsey's study NTX affected self-administration of 0.03 mg cocaine per infusion but not of 0.06 mg per infusion. It may thus be that higher cocaine unit doses mask opioid involvement.

Involvement of opioid systems in effects of cocaine is further supported by intracranial self-stimulation studies [(2) but see (38)] and biochemical studies (15, 18, 32). From the self-administration studies done so far it is not clear if the effects of NTX on cocaine reinforcement are due to action in the brain or to peripheral, less specific effects. In the present study we looked at the effect of intracerebroventricular (ICV) NTX administration on acquisition of intravenous cocaine self-administration. The experimental set-up was the same as the one used in previous studies dealing with systemic NTX treatment (5 days of acquisition, partial food deprivation) (11,25), and a threshold cocaine unit-dose was selected which has previously been shown to be sensitive to acute (11) and chronic (25) systemic treatment with NTX.

METHOD

Details of the apparatus can be found in a previous report (25). Experimentally naive male Wistar rats (TNO, Zeist, Homebred) weighing 220–260 grams at the time of surgery underwent

ABBREVIATIONS

ICV	intracerebroventricular
NTX	naltrexone
DA	dopaminergic
OP	opioid
SI	self-infusions
RL	responses on reinforcement lever
NRL	responses on dummy lever

implantation of a silicon cannula in the jugular vein (37), and at the same time a polyethylene tube was inserted into the right lateral ventricle (3). This tube served to guide a micro syringe. After several days food supply was restricted to achieve a 20% body weight reduction, and the day-night cycle was reversed. Two days later the first of five daily consecutive self-administration sessions began. On each session rats spent a maximum of three hours in operant chambers, which housed two levers. Depression of one lever, marked by a light, triggered a computer controlled infusion pump (FR1), and depression of the other was only registered. During an infusion, 0.25 ml was administered intravenously (containing 30 μg cocaine HCl) through polyethylene tubing in 13 seconds. A session was terminated after either 60 infusions or three hours. Naltrexone or placebo was injected into the ventricle 30 minutes before each session. Subjects received either placebo (saline) or 2 or 5 μg naltrexone in a 2 μl volume, injection of which lasted 10–15 seconds. At the end of the experiment placement of ICV cannulas was checked with methylene blue.

Data were recorded per session and were analysed with analysis of variance for repeated measurements. Three parameters were analysed, being the number of self-infusions (SI), the total number of responses on the infusion or reinforcement lever (RL) which included responses made during infusions, and total number of responses on the dummy or nonreinforcement lever (NRL).

RESULTS

Two experiments were done. In the first, half the animals were treated with 2 μg NTX and half with placebo (saline). In the second experiment 5 μg NTX was administered daily, while control animals received placebo. As no statistically significant differences existed between the two control groups (no main effect of replication nor interaction between replication and time), both experiments were analysed jointly. The data are presented in Fig. 1.

Analysis of variance with repeated measurements revealed a

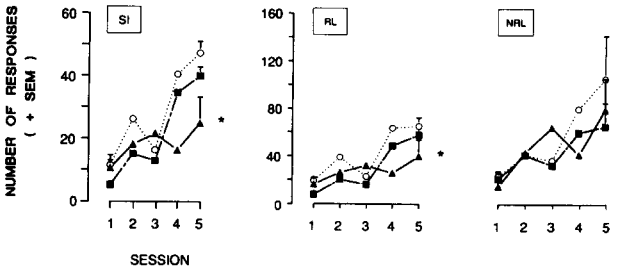

FIG. 1. Effect of ICV NTX treatment on acquisition of intravenous cocaine self-administration in rats. Three parameters are depicted: mean number of self-infusions (SI), of responses on the reinforcement lever (RL) and of responses on the dummy lever (NRL). Subjects were treated daily either with placebo (○, n=12), NTX 2 μg (■, n=6) or NTX 5 μg (▲, n=5). *Different from placebo treatment $p<0.05$ (see text).

main effect of treatment on the SI rate, $F(2,20)=5.05$, $p<0.05$. Subsequently, a multiple comparison test (Scheffe) (29) was done on the SI rate averaged per group over the five sessions (see Table 1). Averaged SI rate differed significantly ($p<0.05$) between the placebo and the 5 μg NTX treated group. The analysis of variance also yielded an overall effect of time, regardless of treatment, $F(4,80)=12.44$, $p<0.001$, but no interaction between treatment and time. Data of the RL parameter parallelled those on SI rate in that main effect of treatment, $F(2,20)=5.31$, $p<0.05$, and of time, $F(4,80)=10.15$, $p<0.001$, were significant, while interaction between time and treatment was not. A multiple comparison test of average RL rate (averaged over five sessions) with the Scheffe procedure showed significant difference ($p<0.05$) between placebo and 5 μg NTX treatment (see Table 1). The number of dummy lever responses (NRL) increased over time, $F(4,80)=4.18$, $p<0.05$, but no differences attributable to treatment were found (see Table 1).

DISCUSSION

In the present experiments placebo-treated rats acquired cocaine self-administration behaviour, as they increased cocaine intake over five self-administration sessions. In the model used, a progressive increase of drug self-infusion over sessions indicates that the drug has rewarding effects [e.g., (8)]. Moreover, the averaged SI rate (over sessions) is contingent upon the unit dose of the drug. With regard to cocaine, the dose-response curve for averaged SI rate is bell-shaped, with an approximately linear relationship in the rising slope. For our purposes the optimal cocaine unit dose was the one that induced a suboptimal

TABLE 1

MEAN NUMBER OF SELF-INJECTIONS (SI), OF RESPONSES ON THE REINFORCEMENT LEVER (RL) AND OF RESPONSES ON THE DUMMY LEVER (NRL) FOR EACH TREATMENT GROUP

Treatment	n	SI	RL	NRL
Placebo	12	142.2 (11.0)	210.3 (16.7)	287.2 (59.5)
2 μg NTX	6	108.0 (9.6)	151.2 (9.9)	221.3 (56.4)
5 μg NTX	5	91.8 (3.8)*	140.8 (5.7)*	244.0 (60.6)

Values in parentheses are standard error of the mean. The data are summed over five sessions. (n: number of cases, NTX: naltrexone).

*Different from placebo, $p<0.05$.

averaged SI rate on the rising slope of the dose-response curve. Using this dose, the averaged SI rate was found to be dose-dependently attenuated by ICV administered NTX. Taken together, the effect of ICV administered NTX in the present study on averaged SI rate mimicks the effect of decreasing the cocaine unit dose.

Naltrexone-treated rats also dose-dependently exhibited lower RL rates than placebo controls, but did not differ in responding on the dummy lever (NRL). The lever-specific difference in responding may reflect reduction of cocaine's reinforcing potency, but nonspecific effects are to be considered. The fact that rats exhibited high response rates on the dummy lever (NRL) (Table 1) suggests that they do not discriminate the source of the infusions. However, previous studies with the present experimental set-up (11,25) showed that NRL responding, in contrast with RL responding, is independent of SI rate. The latter is dependent on cocaine unit-dose. Thus manipulation of reinforcer magnitude either by varying unit-dose or by pharmacological treatment [such as dopamine or opiate antagonists, see (25)] is reflected in SI and in RL rate but not in NRL rate. It should also be noted that the number of operant sessions (five) may be too small for experimentally naive rats to acquire (lever-specific) operant behaviour devoid of nonspecific responding. Although the model used in our study does not by its nature allow for strict dissociation of reinforcing and nonspecific effects (e.g., on locomotor behaviour), the present findings and previous studies indicate that behaviour not related to reward in this model is not affected by the doses of NTX used (11,25). This reasoning is based on absence of an effect of NTX on rate of dummy-lever responding, indicating that locomotor activity was not seriously affected. Moreover, De Vry et al. (11) found an effect of systemic NTX treatment (1 $mg \cdot kg^{-1}$ per session) on acquisition of cocaine self-administration only when a medium cocaine unit dose was used, and not half or double that dose, nor when saline was offered. Thus lever pressing per se is not affected by the dose of NTX used. Schaeffer et al. (27) addressed the matter of nonspecific effect of NTX on locomotor behaviour in detail. They found that 30 $mg \cdot kg^{-1}$ NTX in rats decreased locomotor activity moderately, whereas 1 $mg \cdot kg^{-1}$ NTX affected the response rate for intracranial self-stimulation. Although lower doses of NTX were not tested on locomotor activity, these data do support the notion that NTX affects locomotor behaviour only when given in high doses, whereas effects of NTX on reward-related behaviour occur at much lower doses. Concerning specificity of NTX, other than opioid antagonist actions may take place when given in high doses. NTX has been administered ICV in doses as high as 100 μg (9) before effects could be shown on reward-related behaviour. As NTX is lipophilic, it rapidly diffuses from the ventricles into surrounding brain tissue, possibly leaving little of the initial amount to act upon specific brain systems. In many reports on behavioural effects of ICV treatment with opiate antagonists, quaternary forms are used, because they are less lipophylic and hence do not pass the blood-brain barrier, although the latter may not be quite true (4). In general they are less potent in blocking opioid receptors in vitro than tertiary forms (4). Quaternary NTX has effects on conditioned analgesia (6), on learning (14), on social interaction (21) and on drinking after deprivation (34) in doses between 4 and 10 μg. Vaccarino et al. (35) found that doses as low as 1 μg methylnaloxonium, the quaternary form of naloxone, when administered ICV affected heroin self-administration in rats trained in drug self-administration. Thus, given the finding that quaternary opiate antagonists are less potent but more effective at the site of injection, a dose of 5 μg NTX in the ventricle is neither extremely high nor very low.

It is concluded that blockade of opioid systems in the brain with NTX attenuates the reinforcing effects of cocaine. As discussed above there is little reason to attribute the effect of NTX on SI rate to nonspecific actions. Little is known about the mechanisms by which NTX exerts its effect on cocaine reinforcement. In a variety of studies dopaminergic transmission in the mesocorticolimbic system has been found to be important for the rewarding effects of cocaine (22). Both the DA and the OP systems play a role in reinforcing properties of different drugs (36,40). Interaction between these systems has been suggested to form a neural substrate for reward in general (19,31). Although opioid and dopaminergic systems are known to affect one another in vivo and in vitro (5, 20, 30, 41, 42), the relevance of those findings for reward has not been determined. In addition, the possibility that DA and OP systems operate in independent, parallel, reward mechanisms should however be kept in mind.

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