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A Comparison of Cocaine and its Metabolite Norcocaine: Effects on Locomotor Activity

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ELLIOTT, P J , G M ROSEN AND C B NEMEROFF *A comparison of cocaine and its metabolite norcocaine Effects on locomotor activity* PHARMACOL BIOCHEM BEHAV 26(3) 573–575, 1987 —Intraventricular and intraperitoneal administration of cocaine and its metabolite norcocaine were studied in adult male rats Norcocaine (10–100 μg) had no behavioral activity following central infusion but proved to be toxic at high doses (50–100 mg/kg) when given peripherally Cocaine at high doses (100 μg), produced possible hypoactivity after central injections but produced significant hyperactivity after peripheral administration (20 mg/kg)

Cocaine Norcocaine Locomotor activity Dopamine

COCAINE is a potent central nervous stimulant which has been shown to interact with aminergic neurotransmitter systems. The mechanism(s) involved in the behavioral effects of cocaine are unknown but it is believed that they, at least partly, occur from competitive inhibition of neuronal reuptake of monoamines [1, 6, 7]. Cocaine can diffuse through cell membranes where it is protonated to an ionized form, this ionized species is believed to compete with calcium at sites that control membrane permeability [13,14].

Enzymes present in blood, liver and the central nervous system (CNS) degrade cocaine, producing a variety of metabolites [9, 16, 18]. The first oxidative metabolite of cocaine in the CNS is norcocaine, which can be isolated from brain tissue minutes after systemic administration [8,11] The effects of cocaine and norcocaine on schedule-controlled behavior of pigeons and squirrel monkeys has been studied [15] Peripheral injections of cocaine elicit a large increase in locomotor activity, stereotyped behavior, and in rats with unilateral lesions of the nigro-striatal pathway, rotational activity [4, 5, 12, 14]. However, no behavioral data on central administration of cocaine or infusion of norcocaine are currently available In the present study, therefore, the behavioral effects of both cocaine and norcocaine after peripheral and intracerebroventricular (ICV) administration were investigated.

METHOD

Male Fischer 344 rats (Charles River, Boston, MA) weighing 250–300 g were housed in pairs, in a light- and temperature-controlled room with laboratory chow and water available ad lib. The animals were maintained on corncob bedding to avoid any possible liver-enzyme induction [17]. The rats were anesthetized with Equithesin (1 ml/300 g, IP) and placed in a stereotaxic apparatus (Stoelting Co., Chicago, IL). Subjects were implanted with bilateral guide cannulae (stainless steel, 23 gauge) aimed 1 mm above the lateral ventricle (A–P: −0 4 mm; Lat. ±2 0 mm; Skull· −3 1 mm; Incisor bar +5.0 mm). One week after surgery the animals were placed in photocell cages (43×43×21 cm; Optomex 3/831, Columbus Instruments, Columbus, OH) and allowed to habituate for 1 hr. Groups of 6 rats received either cocaine (10, 50 or 100 μg/side), norcocaine (20, 50 or 100 μg/side) or vehicle (artificial cerebrospinal fluid; CSF) bilaterally in a 5 μl vol./side at a rate of 2 μl/min via 30 gauge needles A one min post-infusion period was used after the drug infusion to allow for diffusion of the drugs from the needle tip.

In a separate experiment, subjects (n=4–6) were administered the drugs intraperitoneally (IP) at the following doses cocaine (20 mg/kg), norcocaine (20, 50 or 100 mg/kg) or vehicle (saline) All IP injection volumes were 1 ml/kg. Following drug injections the animals were returned to their respective cages for 2 hr.

During the test session, locomotor activity (photocell counts) was recorded automatically every 5 min using an on-line computer (PC 900, Columbus Instruments, Columbus, OH) In addition, the subjects were systematically checked (Creese-Iversen scale· [3]) for stereotypy by an un-

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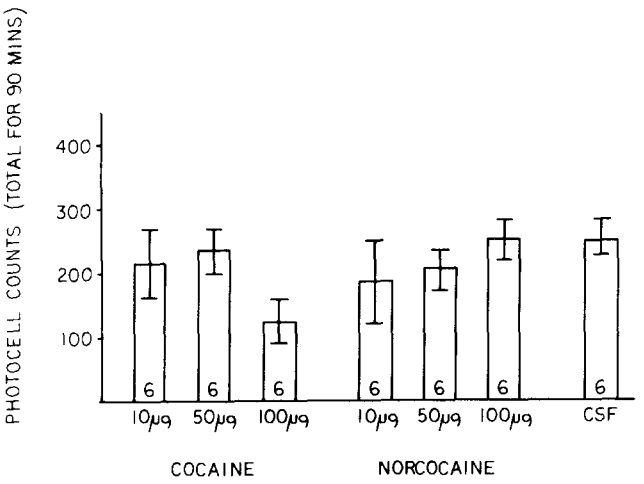

FIG 1 Locomotor activity after intracerebroventricular infusion of norcocaine and cocaine Columns represent group means±S E M Figures in the columns represent the number of subjects per group

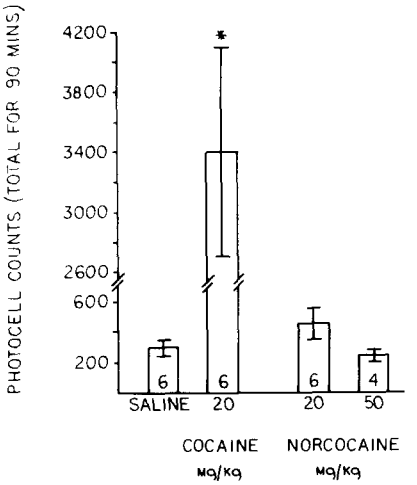

FIG 2 Comparison of the effects of cocaine and norcocaine on locomotor activity after peripheral administration Columns represent group means±S E M Figures in the columns represent the number of animls per group *Indicates $p<0$ 001 with respect to saline

biased observer Each animal was used only once and received one drug injection on each side. Upon termination of the test period the animals were decapitated, their brains removed and later sectioned and stained with cresyl violet to verify cannulae placement; all injection cannulae were found to have broken the corpus callosum and thus the placements were deemed to be correct The data were analysed using analysis of variance and a subsequent post-hoc Students Newman-Keuls test for multiple comparisons. Two tailed significance tables were used in conjunction with the post-hoc test and a probability of $p<0.05$ was considered to be significant. Norcocaine was synthesized using the procedures described previously [2] and used as the acetate salt Cocaine HCl was purchased from Mallinkrodt (St. Louis, MO) and artificial CSF was prepared using the formula from Merlis [10]

RESULTS

For the first five minutes after ICV injection both cocaine (50 and 100 μg) and norcocaine (10, 50 and 100 μg) did increase locomotor activity (data not shown) However, neither ICV administration of cocaine nor norcocaine produced any hyperactivity over the 1½ hr test session In fact, the highest dose (100 μg) of cocaine produced a trend, $F(3,21)=1.94$, $p=0$ 15, towards a reduction in motor activity, when compared to CSF (Fig 1) In contrast, IP administration of cocaine produced a highly significant, $F(2,15)=38.5$, $p<0$ 001, increase in motor activity when compared to vehicle controls and norcocaine (Fig. 2). After peripheral injection of norcocaine, however, the two higher doses (50 and 100 mg/kg) proved to be toxic with 50 and 100% mortality respectively occurring in each group. The subjects exhibited clonic-tonic seizures within 90 sec of the drug infusion and died approximately 2 min later

During the ICV administration the rats infused with norcocaine appeared to be slightly more 'activated' than either the cocaine or the CSF-infused animals However, this behavioral activation was short-lived, and all the drug-treated animals showed similar types of behavior when returned to their respective photocell cages After moving slowly from the center of the cage the rats assumed a hunched posture, usually in a corner No signs of any stereotyped behavior were noted in any of the test animals, i e , sniffing, licking, gnawing, biting. The rats receiving norcocaine or cocaine spent most of the test period in a frozen-like posture, with their eyes open staring at the side or corner of the cage, no other unusual types of behavior were observed At 90 min after the drug administration (ICV) all the subjects were asleep; the CSF animals usually went to sleep 15–20 min after the drug infusion. Rats treated with IP norcocaine appeared to act in an identical fashion to those injected with saline. In contrast, rats given IP cocaine exhibited a marked increase in locomotor activity, $F(2,15)=38$ 5, $p<0$ 001, which persisted for 70 min No stereotypic behavior was observed in any test group and all animals were asleep by 90 min post-peripheral injection

DISCUSSION

The present data confirms previous reports [12,14] that systemic cocaine elicits increased locomotor activity in rats Moreover, the results show, for the first time, behavioral data from direct CNS (i e , ICV) infusions of cocaine and its major oxidative metabolite, norcocaine The data from our experiments actually show a paradoxical effect of cocaine at high (100 μg) ICV doses of the drug there appeared to be a trend towards decreasing motor activity This hypoactivity could possibly arise from cocaine's effect on CNS respiratory centers, which are readily accessible from the CNS ventricular system. Such an action of cocaine has previously been reported [19] Finally, preliminary data from our laboratory (Cain and Nemeroff, unpublished results) indicate that cocaine can in fact be fatal when administered intracisternally

in high doses (≥200 μg). In such studies, animals also exhibited types of behavior similar to those reported in the present experiments. The rapid occurrence of the toxic effect of norcocaine, after peripheral administration, is likely to occur from similar sites of action in the CNS and/or effects on the cardiovascular system

It is known that IP cocaine administration produces an increase in locomotor activity and an increase in dopamine metabolism in the nucleus accumbens [11] These results are usually indicative of mesolimbic dopamine-pathway activation; therefore the failure to increase locomotor activity after ICV infusion of cocaine is puzzling However, the exact mechanism by which systemic cocaine increases motor activity is unclear and furthermore, the distribution of cocaine (and norcocaine) after ICV injections is unknown. It is possible that these drugs might rapidly pass out of the CNS ventricular system into the brain's vascular system, where they are further metabolized, though their lipophyllic nature would argue against this hypothesis. Thus it is uncertain if cocaine can pass from the CNS ventricular system to the sites where it presumably acts upon the mesolimbic dopamine systems, though the present results suggest that this is not the case.

Because neither central nor peripheral administration of norcocaine increased locomotor activity, it is therefore reasonable to conclude that under our behavioral testing conditions, it is inactive

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