Pharmacology Biochemistry & Behavior, Vol. 17, pp. 107–109, 1982. Printed in the U.S.A.

Opiate Effects on Plasma Corticosteroids: Relationship to Dysphoria and Self-Administration

R. A. LAHTI AND R. J. COLLINS

CNS Diseases Research, The Upjohn Company, Kalamazoo, MI 49001

Received 4 September 1981

LAHTI, R. A. AND R. J. COLLINS. *Opiate effects on plasma corticosteroids: Relationship to dysphoria and self-administration.* PHARMAC. BIOCHEM. BEHAV. **17**(1) 107–109, 1982.—Narcotic analgesics have been shown to raise plasma corticosteroids in the rat. This effect has been interpreted as a response of the rat to the dysphoric properties of the drug. This interpretation is based on the observation that drugs such as nalorphine or cyclazocine which cause dysphoria in man elevate plasma corticosteroids in the rat at relatively low doses. Drugs like morphine or pentazocine which induce little dysphoria in man elevate plasma corticosteroids in the rat only at much larger doses. The corticosteroid-elevating effect is mediated by an opiate receptor since naloxone antagonizes the effect of morphine or the analgesic, U-50,488. Those narcotic analgesics which increased corticosteroid levels at low doses were also found to be poorly self-administered by rats. In contrast, compounds like morphine, pentazocine and the analgesic, U-49,274A, which were self-administered at high rates, elevated corticosteroids only after large doses. The relationship between the dose of a drug which causes elevations in corticosteroid levels and whether or not the drug is self-administered further supports the premise that elevated corticosteroid levels induced by analgesics is due to their dysphoric properties.

Corticosteroids Opiate analgesics Dysphoria Self-administration

CHANGES in rat plasma corticosteroids can readily be brought about by environmental stimuli [4] or by the administration of certain drugs [5, 6, 7]. In the case of environmental stimuli, the increase in corticosteroids is, in all probability, due to anxiety associated with the new situation. This anxiety or stress-induced increase in corticosteroids can be reduced by prior administration of antianxiety agents [4]. The elevation in corticosteroids by drugs may be due to several factors, such as a direct adrenal or pituitary action, or by altering feedback systems which control plasma corticosteroid levels. Drugs may also induce a stress response and thereby act in a manner like external stimuli. A possible example of a drug causing an increase in corticosteroids because of stress was the reported finding [5] with pentobarbital. At low doses it reduced the corticosteroid-elevating effect of external stimuli, that is, it had antianxiety activity. Non-stressed rats given large doses had elevations in corticosteroid levels such that the drug appeared to induce stress. Finally, anesthetic doses of pentobarbital resulted in a decrease in corticosteroid levels. Presumably at anesthetic doses, the rat is no longer aware of the large dose-induced sedative effects which may be dysphoric and consequently its corticosteroid levels are low.

Previous studies have shown that opiates also have effects on the pituitary-adrenal axis such as depletion of adrenal ascorbic acid [3] and elevation of plasma corticosteroids in rats [6]. Further, it has been shown that chronic administration of morphine results in adrenal hypertrophy [9]. It has also been demonstrated that rat plasma corticosteroids are elevated following the administration of, and during withdrawal from, levorphanol [2].

A difficulty often encountered in the elevation of narcotic analgesics of the agonist-antagonist type is determining whether or not the agents will produce dysphoria at therapeutic doses, such as happens with cyclazocine and nalorphine. It was considered that the plasma corticosteroid response of rats to chemical agents may be a useful indication of this parameter. This report describes the changes in plasma corticosteroids brought about by narcotic analgesics in the rat. We propose that changes induced by these drugs in the rat may be a useful measure of their capacity to induce dysphoria in man.

METHOD

Male Upjohn Sprague-Dawley rats, weighing approximately 240 grams, were used in all corticosteroid experiments. Rats were housed 5 to a cage for several days before the day of the experiment and given free access to water and food. On the day of the experiment the rats were injected IP with vehicle or drugs at various dose levels and blood collected one hour later via heart puncture under halothane anesthesia. Corticosteroids were determined as previously described [4]. Five rats were used per treatment schedule. Plasma corticosteroid levels in unstressed vehicle-treated rats had a mean value of 16.4±4.8 μg/ml over 7 experiments. ED_{300} values are defined as the dose of drug which would cause corticosteroids to increase to 300% of control

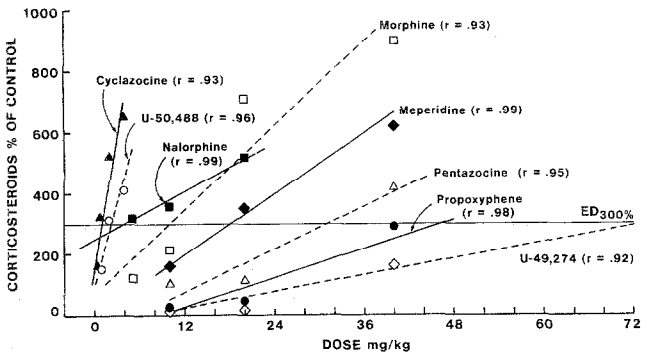

FIG. 1. Effect of various narcotic analgesics on rat plasma corticosteroids. Drug or vehicle was administered IP one hour prior to bleeding via heart puncture. Five rats were used per determination. Control mean values were 16.4±4.8 μg/ml (±S.D.).

TABLE 1

ANTAGONISM OF U-50,488E AND MORPHINE EFFECT BY NALOXONE

Treatment	Corticosteroids μg/100 ml (±S.D.) Without Naloxone	With Naloxone
—	4.9 ± 2.0	4.9 ± 2.9
U-50,488E 2.0 mg/kg	30 ± 14.9*	11 ± 3.0†
Morphine 25.0 mg/kg	57 ± 30.4*	11 ± 10.4†

Naloxone (2.5 mg/kg) was given IP 30 minutes before drug or vehicle and the rats were bled 60 minutes after drug.

*Significantly different from control at $p<0.01$.

†Significantly different from group without naloxone at $p<0.05$.

values. The values were calculated using linear regression analysis. Statistical analysis was performed using Student's *t*-test.

The self-administration procedure has been described previously [1, 12, 13]. Female Sprague-Dawley rats (250–400 grams), implanted several days before use, were placed in the operant chambers. They received a 1 mg/kg dose of test drug with each lever press (FR-1) for 5 days. The dose of test drug was reduced to 0.1 mg/kg/lever response for an additional 5 days. The reinforcement schedule was increased to FR-2 for 2 days and FR-4 for the final 2 days. Only the FR-4 data are reported in the present study. Six naive rats were tested for each drug.

RESULTS AND DISCUSSION

The data presented in Fig. 1 show the effect of various narcotic analgesics on rat plasma corticosteroids (CS) levels. Cyclazocine was the most active compound tested in causing an elevation in CS, and was closely followed in potency by U-50,488 (trans-3,4-dichloro-N-methyl-N-[2-(l-pyrrolidinyl) cyclohexyl-l]-benzeneacetamide), a novel type of opiate analgesic [11], and then nalorphine. These three agents caused CS to increase to 300% of control (ED_{300}) at doses of 0.97, 2.32 and 3.26 mg/kg, respectively. Drugs which had considerably higher ED_{300}'s were morphine (9.97 mg/kg), meperidine (17.7 mg/kg), pentazocine (30.8 mg/kg), propoxyphene (43.5 mg/kg), and a structurally different opiate analgesic [10], U-49,274A (m-[8-(butylmethylamin)-1,4-dioraspiro[4,5]-dec-8-yl]phenyl, monohydrochloride) was effective at 61.4 mg/kg.

To show that the CS elevating effect described above was through an opiate mechanism, an attempt was made to block the activity by pretreatment with the opiate antagonist, naloxone. The results of this study are presented in Table 1. It can be seen that morphine, at 25 mg/kg, and U-50,488, at 2 mg/kg, caused large increases (>600% of control) in CS. When rats were pretreated with 2.5 mg/kg of naloxone, the CS elevating effect was significantly attenuated.

Additional support for the specificity of the CS elevating effect by this class of compounds was demonstrated using U-50,488. This compound, which is a racemic mixture, was separated into its d- and l-enantiomers. The l-enantiomer is an analgesically active species, whereas the d- is not (unpublished observation). When tested for their effect on CS, the l-enantiomer's activity closely paralleled that of the dl- mixture, while the analgesically inactive d-enantiomer was inactive (Table 2). These results point out that the proper stereospecificity is needed along with the opiate activity.

The CS elevating action of the analgesics had an inverse relationship to their self-administration properties. That is, an increase in the dose which elevated plasma corticosteroids to 300% of control (ED_{300}) was linearly related to an increase in the number of lever presses/day (lp/d) for a dose of 0.1 mg/kg/injection on a FR-4 reinforcement schedule (Fig. 2). Compounds such as U-49,274, pentazocine and propoxyphene promoting a rate of lever pressing of from 1,900–3,300 lp/d did not cause elevations of plasma corticosteroids until doses of 30 mg/kg or more were administered. Morphine represents an intermediate drug which had a CS ED_{300} of 9.97 mg/kg, and induced a lever pressing rate of 286 responses/day. Cyclazocine, U-50,488 and nalorphine caused significant elevations of CS at doses of less than 5 mg/kg. Each of these drugs promoted only low levels of lever pressing activity (7–129 lp/d). Of these three drugs, only the response rate for nalorphine was greater than the response rate for rats offered saline (10.6 lp/d).

Linear regression analysis was carried out on the data presented in Fig. 2. A correlation coefficient of r=0.91 ($p<0.01$) was obtained, which indicates a strong correlation between the ED_{300} for CS elevation and the rate of lever pressing. The previous assumption that a low ED_{300} for CS elevation was indicative of dysphoria is supported by the self-administration data; drugs which elevate CS at relatively low doses are not highly self-administered.

The present results suggest that stress or dysphoria induced by opiate analgesics is the common denominator relating corticosteroid levels and self-administration. This stress induces a physiological response, part of which is the elevation of plasma corticosteroids, and a behavioral response which is the avoidance of injections of the stress-promoting drug. Implied in this concept is the idea that reinforcement of lever pressing by opiate analgesics is the sum of its dysphoric and euphoric properties. Drugs such as pentazocine, propoxyphene and U-49,274A which induce stress only at larger doses are self-administered at high rates. Conversely, when drugs cause stress at low doses, they are not avidly self-administered. Since the euphoric and dysphoric properties of opiate analgesics cannot be separated, the concept remains untested. Co-administration of the narcotic

TABLE 2

EFFECT OF ENANTIOMERS OF U-50,488 ON PLASMA CORTICOSTEROID

Treatment	Dose (mg/kg)	Corticosteroids μg/100 μl (±S.D.)
Vehicle	—	23.2 ± 16
(dl)	1.0	36.6 ± 30
(dl)	2.0	72.5 ± 45.7
(dl)	4.0	100 ± 8.2*
(l)	1.0	67 ± 16.8*
(l)	2.0	81 ± 30.7*
(l)	4.0	114 ± 18.6*
(d)	1.0	14.3 ± 7.6
(d)	2.0	14.5 ± 8.6
(d)	4.0	13.6 ± 5.2

Five rats per group.
*Significantly different from control at $p<0.01$.

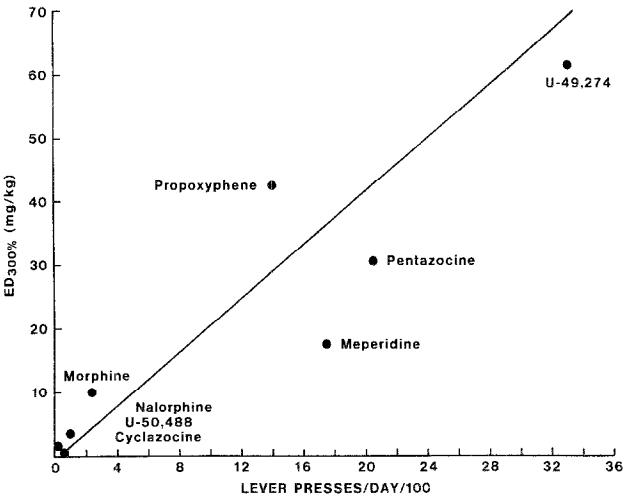

FIG. 2. Correlation of ED_{300} for elevating plasma corticosteroids and rate of self administration. Rate of self administration on FR-4 schedule with a dose of 0.1 mg/kg/injection was utilized. Correlation coefficient was r=0.91 and $p<0.01$.

antagonist, naloxone, with a reinforcing opiate abolishes both the corticosteroid-elevating effect and also abolishes its ability to promote lever pressing. The relationship between self-administration properties of a drug and its effect on plasma corticosteroids appears useful. Effects of drugs on plasma corticosteroids can be measured easily, whereas self-administration studies require animals with indwelling cannulae and chronic drug dosing.

The converse situation, that drugs which do not increase plasma corticosteroids at low doses will necessarily be strong reinforcers, may not be the case. Other important criteria may be involved in the reinforcing properties of drugs.

The conclusion derived from these studies is that the effects of opiate analgesics on plasma corticosteroid levels may be used as an indicator of the aversiveness or dysphoria of this class of drugs. Compounds which cause significant elevations in CS at doses near the effective analgesic dose may be too dysphoric for use. Cyclazocine and nalorphine, which are dysphoric in humans and were shown to cause large elevations in CS at relatively low doses, support this interpretation.

ACKNOWLEDGEMENTS

The following pharmaceutical companies are acknowledged for generously supplying drugs: Winthrop, Eli Lilly, and Endo.

C. Barsuhn is acknowledged for his excellent technical assistance and S. J. Crowder for skillful manuscript preparation.

REFERENCES

1. Collins, R. J., J. R. Weeks and P. I. Good. Evaluation of the reinforcing properties of psychoactive drugs using rats. *Proceedings, Committee on Problems of Drug Dependence.* 1978, pp. 510 and 521.
2. Eisenberg, R. M. and S. B. Sparber. Changes in plasma corticosterone levels as a measure of acute dependence upon levorphanol in rats. *J. Pharmac. exp. Ther.* **211:** 364–369, 1979.
3. George, R. and E. Leong Way. Studies on the mechanism of pituitary-adrenal activation by morphine. *Br. J. Pharmac.* **10:** 260–264, 1955.
4. Lahti, R. A. and C. Barsuhn. The effect of minor tranquilizers on stress-induced increases in rat plasma corticosteroids. *Psychopharmacologia* **35:** 215–220, 1974.
5. Lahti, R. A. and C. Barsuhn. The effect of various doses of minor tranquilizers on plasma corticosteroids in stressed rats. *Res. Communs chem. Path. Pharmac.* **11:** 595–603, 1975.
6. Lahti, R. A. and C. Barsuhn. The effect of antidepressants on L-5-HTP-induced changes in rat plasma corticosteroid. *Res. Communs chem. Path. Pharmac.* **28:** 343–349, 1980.
7. Lahti, R. A. and C. Barsuhn. Benzodiazepines, stress and rat plasma corticosteroids: The role of indoleamines. *Res. Communs chem. Psychol. Psychiat Behav. 5:* 369–383, 1980.
8. Nikodijevic, O. and R. P. Maickel. Some effects of morphine on pituitary-adrenocortical function in rat. *Biochem. Pharmac.* **16:** 2137–2142, 1967.
9. Tanabe, T. and E. J. Cafruny. Adrenal hypertrophy in rats treated chronically with morphine. *J. Pharmac. exp. Ther.* **122:** 148–153, 1958.
10. VonVoigtlander, P. F., D. Lednicer, R. A. Lewis and D. D. Gay. *Endogenous and Exogenous Opiate Agonists and Antagonists.* New York: Pergamon Press, 1980, p. 17.
11. VonVoigtlander, P. F., R. J. Collins, R. A. Lewis and G. L. Neff. U-50,488 (trans-3,4-dichloro-N-methyl-N-[2-(l-pyrrolidinyl) cyclohexyl]-benzeneacetamide): Prototype of a new class of narcotic analgesics. *Pharmacologist* **23:** 112, 1981.
12. Weeks, J. R. Experimental morphine addiction: Method for automatic intravenous injections in unrestrained rats. *Science* **138:** 143–144, 1962.
13. Weeks, J. R. and R. J. Collins. Self-administration of morphine in the rat: Relative influence of fixed ratio and time-out. *Pharmac. Biochem. Behav.* **9:** 703–704, 1078.