Pharmacology Biochemistry & Behavior, Vol 29, pp 609–612 © Pergamon Press plc, 1988 Printed in the U S A 0091-3057/88 $3 00 + 00

Effects of Mu Receptor Agonists and Droperidol on Motor Coordination in Mice

M. BANSINATH, J. S FISHER, C. K. TANG, H. TURNDORF AND M. M. PUIG[1]

Department of Anesthesiology, NYU Medical Center 550 First Avenue, New York, NY 10016

Received 13 July 1987

BANSINATH, M , J S FISHER, C K TANG, H TURNDORF AND M M PUIG *Effects of mu receptor agonists and droperidol on motor coordination in mice* PHARMACOL BIOCHEM BEHAV **29**(3) 609–612, 1988 —The effects of morphine, fentanyl, sufentanil and droperidol on motor coordination in mice were studied Animals were trained to complete successfully the rotarod test before assessing the effects of drugs Administration of analgesic doses of the mu agonists morphine, fentanyl and sufentanil did not inhibit motor coordination Droperidol produced a dose related inhibition of motor coordination When a subthreshold dose of droperidol was administered followed by an opiate, a significant inhibition of motor coordination was observed The results indicate that although analgesic doses of mu opioid agonists do not affect motor coordination, their combination with droperidol results in motor incoordination The mechanisms and/or opioid receptor sub-types involved in this in vivo interaction remain to be established

Opiates Opioid receptors Mu receptor agonists Motor coordination Droperidol

DOPAMINERGIC mechanisms are known to be involved in modulation of motor activity [17]. In vitro evidence indicates that morphinans and benzomorphans bind to dopaminergic sites in the striatum [5] and in vivo, the dopamine receptor blocker spiperone has been shown to decrease morphine induced hypermotility [11] β-Endorphin has been reported to induce a pattern of behavior similar to that of neuroleptic drugs [13] Furthermore, the neuroleptics like haloperidol and droperidol have been shown to release endogenous opioids [18,22] The opiate antitussive codeine, a weak mu opioid receptor agonist, alters visuo-motor coordination and dynamic visual acuity [3]

To induce neuroleptanesthesia, extremely potent mu opioid receptor agonists like fentanyl and sufentanil are often combined with the neuroleptic droperidol Droperidol is a dopamine receptor antagonist [4] and has a longer duration of action when compared to fentanyl [16] Hence, due to residual concentrations of droperidol and opiates in vivo during neuroleptanesthesia, droperidol is likely to modulate opioid induced changes in motor coordination. The present study was aimed to characterize the motor coordination changes induced by analgesic doses of opioids combined with droperidol The rotarod test—a model to test motor coordination in rodents [23]—was used to assess the effect of combinations of fentanyl, sufentanil and morphine with droperidol on motor coordination in mice. The results indicated a positive in vivo drug-interaction between the opiates and droperidol

METHOD

Male Swiss Webster mice weighing 25–30 g (Taconic Farms, PA) were housed five per cage in a room with controlled temperature (22±2°C), humidity and artificial light (06·30–19 00 hr). The animals had free access to food and water and were used after a minimum of four days acclimation to the housing conditions

Droperidol, fentanyl citrate, sufentanil citrate (Janssen Pharmaceutica, Piscataway, NJ) and morphine sulfate (Mallinckrodt, Inc , St Louis, MO) were dissolved in deionized water just before use. All drugs were injected SC in a volume of 10 ml/kg The effects of droperidol (0 5, 1, 2 5 and 5 mg/kg), morphine (1, 2 and 3 mg/kg), fentanyl (5, 25 and 50 μg/kg) and sufentanil (1, 5 and 10 μg/kg) and of the same doses of opiates in combination with droperidol (0 5 mg/kg) were assessed in groups of six mice Animals in the control group received isotonic saline injections Droperidol was injected 60 minutes prior to and the opiates 30 (morphine) or 15 (fentanyl and sufentanil) minutes prior to testing based on peak times established in previous analgesic tests in our laboratory [20]

The details of the rotarod test procedure were essentially similar to those published earlier [1]. Animals were trained to perform the task before testing the drug effects A rotarod of 2 75″ in diameter with partitions dividing the rod into three equal positions and with a rotation speed of 15 revolutions per minute was used A stay on the rod for a maximum

[1]Requests for reprints should be addressed to Dr M M Puig, Department of Anesthesiology, NYU Medical Center, 550 First Avenue, New York, NY 10016

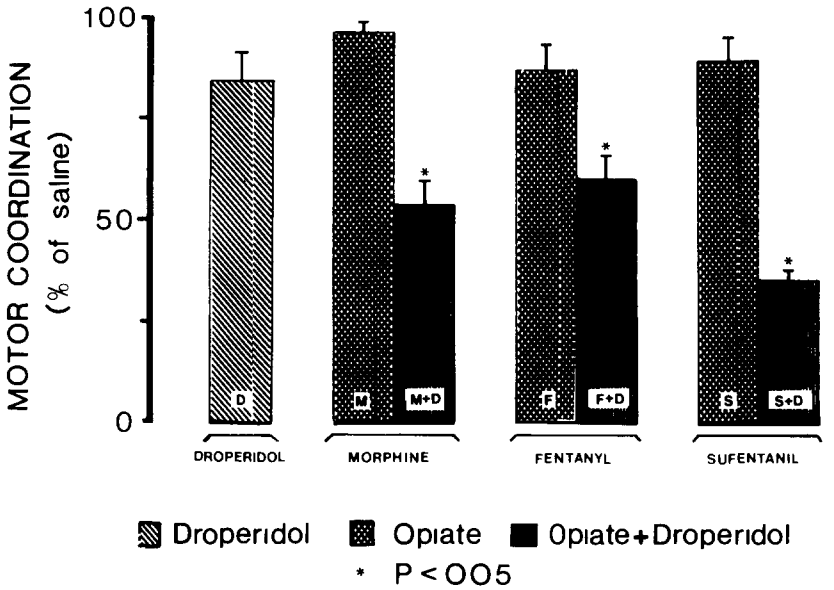

FIG 1 Effects of opiates and droperidol on motor coordination in trained mice The animals were trained for five days to complete the rotarod test The time of stay on the rod for each mouse was converted to percent of mean stay of the saline injected group Mean±SEM is represented (n=6) Morphine (3 mg/kg), fentanyl (50 μg/kg) or sufentanil (10 μg/kg) alone did not affect motor coordination The same dose of opiate combined with droperidol (0 5 mg/kg) significantly inhibited motor coordination ($p<0$ 05) Bar labelled droperidol represents the effect of 0 5 mg/kg droperidol alone *$p<0$ 05 when comparing the results between the opiate alone and the opiate-droperidol combination using a two-tailed Student's t-test

period of 90 sec was taken as the criterion for the successful completion of the test Animals were subjected to two training sessions per day, with two trials spaced four hours apart on each of five consecutive days Animals which could not be trained to complete successfully the test by the ninth trial were not included in the study The tenth trial was preceded by saline or drug(s) administration(s) Each group with a minimum of six animals per group was tested an equal number of times in three rod positions. Group-wise data of the stay on rod in seconds was recorded

The time of stay on the rod for each animal was converted to percent of the mean time of the saline treated group Prior to subjecting the data to statistical analysis, individual percentages as a measure of motor coordination were converted to their square roots [10,23] The data were subjected to ANOVA followed by Student's t-test A p value of <0 05 was considered as criterion for significance

RESULTS

Droperidol induced significant dose related inhibition of motor coordination ($p<0$ 05) The percents motor coordination in droperidol treated groups were 87±6, 57±5, 49±6 and 40±3% of the saline control for groups which received 0 5, 1, 2 5 and 5 mg/kg of droperidol, respectively The results were significant for groups which received 1, 2 5 and 5 mg/kg of droperidol ($p<0$ 05) when compared to the saline treated controls However, 0 5 mg/kg of droperidol did not significantly affect motor coordination The effects of morphine, fentanyl and sufentanil on motor coordination were determined in groups of six mice per dose The doses of opiates used were based on their ED_{50}'s in two analgesic tests in our laboratory [20] Group-wise data of the mean duration of stay on the rod in seconds (mean±SEM) after different doses of opiates alone and in combination with droperidol (0.5 mg/kg) are shown in Table 1 No significant inhibition of motor coordination was observed for morphine, fentanyl or sufentanil when the animals received the opiates alone

TABLE 1

DROPERIDOL NARCOTIC INTERACTION IN THE ROTAROD TEST IN MICE

Group	Dose (μg/kg SC)	Stay on Rod in Seconds—mean ± SEM Control (a)	Test (b)
Saline		89 0 ± 0 9	77 5 ± 5 3
Morphine	1000	89 5 ± 0 5	65 2 ± 6 7*
	2000	85 0 ± 3 0	52 3 ± 4 8*
	3000	83 7 ± 3 8	47 0 ± 5 0*
Fentanyl	5	79 0 ± 3 9	79 3 ± 5 7
	25	75 5 ± 5 2	62 0 ± 3 4
	50	75 8 ± 6 1	52 2 ± 5 7*
Sufentanil	1	84 7 ± 3 2	76 7 ± 4 4
	5	76 3 ± 6 0	62 3 ± 7 9
	10	78 0 ± 5 5	31 3 ± 2 0*

(a) Group with single drug treatment (Saline or opiate)
(b) Group with combination of drugs—0 5 mg/kg droperidol and the opiate
*Significantly different ($p<0$ 05) when compared with respective control (a) group

However, when a subthreshold dose of droperidol (0 5 mg/kg) was combined with the opiates, motor incoordination was observed with morphine, fentanyl and sufentanil (Table 1) The data for the highest dose of opiate alone and in combination with droperidol expressed as percent of saline control are presented in Fig 1 Groups which received the opiate and droperidol combination treatment showed significant motor incoordination as compared to the respective opiate treated groups ($p<0$ 05) Catalepsy was not observed in animals treated with (1) droperidol alone (2) opiates alone or (3) the combination of droperidol (0 5 mg/kg) with analgesic doses of opiates.

The results indicate that mu receptor agonists in analgesic doses do not inhibit motor coordination in mice trained for the testing procedure. However, the combination of a subthreshold dose of droperidol with mu-opioid agonists induces motor incoordination in mice in the rotarod test

DISCUSSION

In general, central nervous system depressants reduce while stimulants enhance the rotarod performance of mice [15] Behavioral effects of opioids are known to be species dependent [2]. Opiate induced changes in motor activity of rats and mice have been often reported using activity cages [6, 14, 21]. Only recently, the rotarod test has been used to assess the specific roles of the opiate receptor sub-types in mediating the sedative effects of opiates [8, 9, 11] In mice and rats which are not trained prior to testing on the rotarod, mu as well as kappa agonists have been shown to decrease latency [8,9] However, it is important to emphasize that morphine induced changes in motor activity cannot be observed in mice which are pre-exposed to the test environment [19] In rats trained on an accelerating rotarod, only kappa (ketocyclazocine) but neither mu (morphine) nor sigma (SKF-10,047) agonists affected the rotarod performance [11] The results of the present study with a rotarod of fixed rotations and trained mice indicate that mu agonists in the analgesic doses do not inhibit motor coordination.

Few laboratories have reported the behavioral interaction of droperidol with opiates In what appears to be a single report to date, based on a subjective assessment, droperidol was reported not to augment central nervous system depression induced by fentanyl in mice [24] However, our results, using an objective assessment of the interaction, indicate that opiate-droperidol combination affects motor coordination

Variable motor response to opioids in mice has been explained as being likely due to (1) opiate receptor sub-types, mu and sigma receptors stimulating while kappa receptors depressing motor activity (2) distinct dopaminergic mechanisms mediating mu, sigma and kappa opiate effects [7,12] Hence, droperidol induced blockade of dopaminergic sites could be considered as the mechanism of the observed interaction between droperidol and opiates Droperidol is also known to potentiate the analgesic effect of fentanyl and sufentanil but not of morphine [20,24] Such qualitative differences between mu agonists was not observed in the present results on motor coordination The dichotomy with reference to the analgesic versus sedative effects of the mu agonists has been explained on the basis of involvement of different receptors [3,9] Accordingly, the disparity in the results on the motor coordination versus analgesic effects suggests the possibility that the site and/or the mechanisms mediating the interaction are not the same for analgesic and sedative effects

REFERENCES

1 Bansinath, M , A C Bose, S Hema and M N Guruswami Interaction of metamizol with some hypnotics in rats *Arch Int Pharmacodyn* **260:** 14–27, 1982

2 Booth, N H Neuroleptanalegesics, narcotic analgesics and analgesic antagonists In *Veterinary Pharmacology and Therapeutics,* edited by N H Booth and L E McDonald Ames, IA Iowa State University Press, 1982, pp 267–296

3 Bradely, C M and A N Nicholson Effect of a mu-opioid receptor agonist (Codeine phosphate) on visuo-motor coordination and dynamic visual acuity in man *Br J Clin Pharmacol* **22:** 505–512, 1986

4 Burt, D R , I Creese and S H Snyder Properties of (3H) haloperidol and (3H) dopamine binding associated with receptors in calf brain membranes *Mol Pharmacol* **12:** 800–812, 1976

5 Czlonkowski, A , V Hollt and A Herz Binding of opiates and endogenous opioid peptides to the neuroleptic receptor sites in the corpus striatum *Life Sci* **22:** 953–962, 1978

6 Fidecka, S , D Malec and R Langwinski Central action of narcotic analgesics II Locomotor activity and narcotic analgesics *Pol J Pharmacol Pharm* **30:** 5–16, 1978

7 Goodman, R R and G W Pasternak Multiple opiate receptors In *Analgesics Neurochemical, Behavioral, and Clinical Perspectives,* edited by M Kuhar and G Pasternak New York Raven Press, 1984, pp 69–96

8 Hayes, A G and M B Tyers Determination of receptors that mediate opiate side effects in the mouse *Br J Pharmacol* **79:** 731–736, 1983

9 Hayes, G A , M Skingle and M B Tyers Effect of B-funaltrexamine on opioid side-effects produced by morphine and U-50,488H *J Pharm Pharmacol* **37:** 841–843, 1985

10 Irwin, S , M Slabok and G Thomas Individual differences I Correlation between control locomotor activity and sensitivity to stimulant and depressant drugs *J Pharmacol Exp Ther* **123:** 206–211, 1958

11 Iwamoto, E T Locomotor activity and antinociception after putative Mu, Kappa and Sigma opioid receptor agonists in the rat Influence of dopaminergic agonists and antagonists *J Pharmacol Exp Ther* **217:** 451–460, 1981

12 Iwamoto, E T and W R Martin Multiple opioid receptors *Med Res Rev* **1:** 411–440, 1981

13 Jacquet, Y F and N Marks The C-fragment of B-lipotropin An endogenous neuroleptic or antipsychotogen *Science* **194:** 632–635, 1976

14 Kalivas, P W and P Duffy Sensitization to repeated morphine injection in the rat possible involvement of A10 dopamine neurons *J Pharmacol Exp Ther* **241.** 204–212, 1987

15 Kinnard, W J , Jr and N Watzman Techniques utilized in the evaluation of psychotropic drugs on animal activity *J Pharm Sci* **55:** 995–1012, 1966

16 Marshall, B E and H Wollman General anesthetics In *Goodman and Gillman's The Pharmacological Basis of Therapeutics,* 7th edition, edited by A G Gillman, A S Goodman, T W Rall and F Murad New York MacMillan, 1985, pp 276–301

17 Martin, G E and R J Bendesky Mouse locomotor activity An in vivo test for dopamine autoreceptor activation *J Pharmacol Exp Ther* **229:** 706–711, 1984

18 Milanes, M V , M L Vargas and M M Puig Haloperidol treatment increases the biosynthesis and release of endorphins in the guinea-pig ileum *J Pharm Pharmacol* **36:** 446–449, 1984

19 Oliverio, A and C Castellano Experience modifies morphine induced behavioral excitation in mice *Nature* **252:** 229–230, 1974
20 Statile, L , M M Puig, W Warner, M Bansinath, M Lovitz and H Turndorf Droperidol enhances fentanyl and sufentanil but not morphine analgesia *Gen Pharmacol,* in press
21 Tepper, P and J H Woods Changes in locomotor activity and naloxone-induced jumping in mice produced by WIN35,197-2 (ethylketazocine) and morphine *Psychopharmacology (Berlin)* **58:** 125–129, 1978
22 Vargas, M L , J A Martinez and M V Milanes Effects of droperidol on the biosynthesis and release of endogenous opioid peptides in guinea-pig ileum *Gen Pharmacol* **18:** 283–286, 1987
23 Watzman, N , H Barry, III, W J Kinnard, Jr and J P Buckley Influence of certain parameters on the performance of mice on the rotarod *Arch Int Pharmacodyn* **169:** 362–374, 1967
24 Yelnosky, J and J F Gardocki A study of the pharmacologic actions of fentanyl citrate and droperidol *Toxicol Appl Pharmacol* **6:** 63–70, 1964