Pharmacology Biochemistry and Behavior, Vol. 60, No. 2, pp. 519–525, 1998

0091-3057/98 $19.00 + .00

ELSEVIER

PII S0091-3057(98)00002-1

Reversal of Learned Helplessness by Morphine in Rats: Involvement of a Dopamine Mediation

A. BESSON,* A. M. PRIVAT,* A. ESCHALIER† AND J. FIALIP*

*Laboratoire de Pharmacologie, Faculté de Pharmacie,
†Laboratoire de Pharmacologie Médicale, Faculté de Médecine, 28,
Equipe NPPUA (Neuropsychopharmacologie, Université d'Auvergne),
Place Henri Dunant, BP38, 63001 Clermont-Ferrand cedex 1, France

Received 14 August 1997; Revised 12 December 1997; Accepted 12 December 1997

BESSON, A., A. M. PRIVAT, A. ESCHALIER AND J. FIALIP. *Reversal of learned helplessness by morphine in rats: Involvement of a dopamine mediation*. PHARMACOL BIOCHEM BEHAV **60**(2) 519–525, 1998.—The aim of this study was to examine the role of dopamine neurotransmission in the effects of morphine in the learned helplessness paradigm in rats, a generally recognized model of depression. In this model, rats first exposed to inescapable shocks (stressed rats) exhibited an escape deficit in a subsequent shuttle-box test performed 48 h later for 3 consecutive days. The numbers of escape failures and intertrial crossings (motor activity during each intertrial interval) were recorded. Morphine was injected twice daily for 5 days (6 mg/kg/day, SC), and haloperidol, a preferential D_2-dopamine receptor antagonist, was injected IP 15 min before each shuttle-box session. At the highest dose tested (150 μg/kg) haloperidol mimicked the behavioral deficit produced by inescapable shocks. A 37.5 μg/kg dose of haloperidol, which was ineffective by itself, reversed the morphine-induced improvement of escape behavior in previously stressed rats and the morphine-induced increase in intertrial activity in both stressed and nonstressed animals. These results support roles (a) for a dysregulation of dopaminergic neuronal activity in the expression of escape deficit subsequent to an inescapable aversive situation, and (b) for a dopaminergic mediation in the effects of morphine in the learned helplessness paradigm. © 1998 Elsevier Science Inc.

Learned helplessness Morphine Haloperidol Dopamine Rats

IN the last decades, the learned-helplessness paradigm has elicited a great deal of interest as an animal model of depression. In this model, exposure of rodents to an uncontrollable aversive event results in several behavioral alterations including impairments in the learning of a subsequent active escape response, the so-called "learned helplessness" behavior (32,39,40,50), and reduced responsiveness to rewards (52), which are reminiscent of the feeling of distress and the loss of pleasure expressed in depressive patients (3,49). The validity of this animal model is supported by the efficacy of various antidepressants to reverse the observed behavioral alterations (9,33,34). These considerations, together with accumulating evidence that stressful events can help to precipitate depression (5,24), emphasize the relevance of the learned-helplessness paradigm to study the neurochemical basis underlying the behavioral impairments induced by stress in rodents, which are similar to some human depressive symptoms.

Anatomical investigations have shown the presence of both dopamine (DA) and opioid systems in brain areas involved in the control of motivational and emotional processes (2,13,14,41), suggesting their involvement in affective disorders (2,6,10,15,17,48), and in response to stressful events (1,8,12,23,29). Although the role of dopamine and opiates in clinical depression is debated [see (15,17,48) for review], preclinical studies using animal models of depression related to stress point to a decreased activity of these systems of neurotransmission, at least in the expression of certain symptoms, such as anhedonia and feeling of helplessness (7,12,25,40, 42,46,50,51). Moreover, both anatomical and functional interactions exist between the dopamine and opioid systems in the brain; mu-opioid receptors are located on both presynaptic DA neurons and GABAergic interneurons in the ventral tegmental area (VTA) and the substantia nigra (SN) (14,30). Stimulation of mu-opioid receptors increases DA release and

Requests for reprints should be addressed to Prof. J. Fialip, Laboratoire de Pharmacologie, Faculté de Pharmacie, 28, Place Henri Dunant, BP38, 63001 Clermont-Ferrand cedex 1, France.
The procedure used to kill the animals in this study is not allowed in UK or USA.

DA turnover, and the firing of DA neurons in the SN and VTA (14,18,30). From a behavioral standpoint, the rewarding and locomotor effects of morphine are related to DA neuronal activity (26,31,44). Other findings suggest that stress-induced release of endogenous opioids cause an excitation of mesocorticolimbic dopamine neurons. For instance, naloxone or naltrexone antagonized or prevented the increased dopamine metabolism induced by footshocks (22,37) or immobilization (11). Recently, using the learned-helplessness model in the rat, we demonstrated that morphine both reversed the escape deficit induced by footshocks and increased intertrial crossing in helpless rats (7). We therefore undertook to determine whether the antidepressant-like effect of morphine in this model also involved a dopamine mediation. For this purpose we studied whether blockade of dopamine receptors by haloperidol, a preferential D_2-DA receptor antagonist, altered the behavioral effects of morphine, a prototypic mu-opioid receptor agonist, in the learned-helplessness paradigm in the rat.

METHODS

Animals

Male Wistar rats (centre d'élevage, Janvier, France) were used, weighing 190 to 230 g at the beginning of the experiments. They were housed six per cage under standard laboratory conditions, with free access to food and water. All the experiments took place 1 week after the reception of animals.

Apparatus and Procedure

The experimental procedure used below refers to the method described by Martin et al. (33). The experimental protocol was approved by a local ethical committee and complies with the guidelines for animal experimentation of the N.I.H. (1985) and with French legislation (decrees of 20/10/87 and 19/04/88).

Shock pretraining. The rats were divided into two groups. The first group, stressed (S), was exposed to unsignaled, inescapable electric footshocks delivered through a stainless grid floor (1 m × 0.3 m), allowing simultaneous stimulation of several rats. Randomized, scrambled shocks (15-s duration, 0.8 mA, every minute ± 15 s) were generated by an Apelex model "LE 100-26" shocker connected to an Apelex "LI 10-31S" random generator. Stressed animals subjected to inescapable shocks were individually placed in Plexiglas chambers (20 × 10 × 10 cm) onto the electrified grid floor for 1 h. A second group, nonstressed (NS), was placed for 1 h in the same conditions except that the shock generator was turned off. Pretraining was performed on day 1, between 0800 to 1500 h.

Avoidance–escape training. Forty-eight hours after pretraining, all the animals were exposed to an avoidance–escape task in automated two-way shuttle-boxes (OSYS-OrgaSystem) with black walls and an electrified grid floor. Each shuttle-box was divided into two equal-size compartments by a stainless steel partition with a gate that provided access from one compartment to the other. Rats were placed one in each shuttle-box. A 5-min adaptation period was allowed before beginning a series of 30 trials, 24 s apart. In each trial, a light signal came on for the first 3 s (conditioning signal) during which rats were allowed to avoid shock (avoidance response). If no crossing occurred within this period, an electric footshock (3 s duration, 0.8 mA) was delivered. A single crossing from the electrified compartment to the other within this latter period was considered as an escape response. If no escape response occurred, light and shock were discontinued, and an escape failure was recorded. Two parameters were counted: escape failures (EFs) and intertrial crossings (ITCs). The number of EFs was considered as an index of the helpless behavior. ITC is related to the number of crossings in the interval between two trials and could be considered as a measure of a "nonspecific" motor activity (i.e., a motor activity other than the desired escape response) (16). The avoidance–escape session was repeated on days 4 and 5, but no period of adaptation was allowed.

Drug Administration

Two sets of experiments were performed.

Influence of different doses of haloperidol on escape behavior. This series of experiments was performed to determine the dose of haloperidol to be used in further interaction studies.

Rats submitted to inescapable shocks (S) or not (NS) on day 1 were divided in the same manner, as follows (8 to 12 rats/group): (a) control rats received water (S-C and NS-C groups); and (b) six other groups received haloperidol at three different doses: 37.5 μg/kg (NS-H37.5 and S-H37.5); 75 μg/kg (NS-H75 and S-H75); 150 μg/kg (NS-H150 and S-H150). All the haloperidol-treated groups received the drug acutely 15 min before each shuttle-box session on days 3, 4, and 5.

Morphine × haloperidol interaction. A second series of experiments was performed to determine whether the effects of morphine in the learned-helplessness paradigm (7) were dependent on a dopamine receptor activation. We also studied the interaction between haloperidol, a preferential D_2-DA receptor antagonist, and morphine, a mu-opioid agonist, on both EF and ITC.

Rats submitted to inescapable shocks (S) or not (NS) on day 1 were divided in the same manner as follows (8 to 12 rats/group): (a) control rats received water (S-C and NS-C groups); (b) two groups received 6 mg/kg/day of morphine (S-M and NS-M groups). The first dose was injected 4 h after the session of inescapable shocks (S-M) on day 1, and then twice daily (4 mg/kg/day in the morning, and 2 mg/kg/day in the evening) on 4 consecutive days. The morning injections on days 3, 4, and 5 were given 30 min before the shuttle-box session. The dose of 6 mg/kg/day was chosen because previous experiments had shown that 4 to 8 mg/kg/day of morphine completely reversed escape failures induced by inescapable shocks (7); and (c) two other groups were treated with the morphine–haloperidol combination: NS-M-H and S-M-H groups. The 37.5 μg/kg dose of haloperidol was used because it was ineffective by itself in our procedure. Haloperidol was acutely injected in the morning on days 3, 4, and 5, 15 min after morphine injection and 15 min before the shuttle-box test.

Morphine hydrochloride (Coopération Pharmaceutique Française, Melun, France) was dissolved in saline and injected subcutaneously (SC) in a volume of 1 ml/kg. Haloperidol (Janssen-Cilag, Boulogne-Billancourt, France) was dissolved in saline and injected intraperitoneally (IP) in a volume of 2.5 ml/kg. Control rats were injected IP with water.

Statistical Analysis

The number of EFs and ITCs recorded over the 30 trials of each shuttle-box session were expressed as mean ± SEM for each session.

The statistical analyses concerning the EFs were performed by two-way analyses of variance with repeated mea-

sures (RM-ANOVAs). The design comprised two grouping factors ("stress" and "treatment"), and one dependent variable (EF) that was measured three times (days 3, 4, 5). Once the fully factorial RM-ANOVA was estimated, a post hoc Bonferroni test for multiple pairwise comparisons was performed.

Because the individual data concerning ITCs in the second set of experiments did not follow a normal distribution, these data were analyzed by a nonparametric test, the Kruskal–Wallis ANOVA on ranks, followed by the Dunnett test for multiple pairwise comparisons. We used Spearman's rank correlation to detect any possible association between the two parameters (ITC and EF) in NS-M, S-M, NS-MH, and S-MH groups, the scores obtained during the three shuttle-box tests being pooled for each type of treatment.

In all cases, the 0.05 level of significance was chosen.

RESULTS

Effects of Haloperidol in the Learned Helplessness Model (Fig. 1)

The overall RM-ANOVA revealed a significant effect of the factors "stress," $F(1, 68) = 15.47$, $p < 0.05$, and "treatment," $F(3, 68) = 5.46$, $p < 0.05$, as well as a slight but significant "stress" × treatment interaction, $F(3, 68) = 2.84$, $p = 0.044$. More precisely, as shown in Fig. 1, the multiple pairwise comparisons revealed a significant impairment of escape response in nonstressed rats injected acutely with 75 or 150 μg/kg of haloperidol, 15 min before the shuttle-box session (NS-H75 vs. NS-C and NS-H150 vs. NS-C: $p < 0.05$), whereas no dose of haloperidol induced any significant change in the escape behavior in previously stressed rats. The multiple pairwise comparisons showed that exposure of nontreated rats to IS significantly impaired their escape response in a subsequent escape task compared with their nonstressed counterparts, whatever the day of the shuttle-box session (S-C vs. NS-C: $p < 0.05$, days 3, 4, 5). Interestingly, the "within-subject" analysis did not reveal any significant influence of the repeated measures on factors "stress" and "treatment."

Effects of Haloperidol on Responses to Morphine in the Learned Helplessness Paradigm (Fig. 2).

Escape behavior (Fig. 2A). The overall RM-ANOVA revealed a significant effect of the factors "stress," $F(1, 83) = 36.62$, $p < 0.05$, and "treatment," $F(3, 83) = 5.43$, $p < 0.05$, but no interaction between these two factors, $F(3, 83) = 2.43$, $p > 0.05$. Moreover, the "within-subject" analysis revealed that the effect of stress varied with the repetition of the escape sessions in a nonlinear way (multivariate repeated-measures analysis for "repeated measures × stress") $F(2, 82) = 4.06$, $p < 0.05$. This may be because the number of EFs tended to decrease between the first and the third shuttle-box sessions in the NS-C group while it tended to increase in the S-C group. The multiple pairwise comparisons showed that exposure of the rats to IS significantly impaired their escape response during the three subsequent shuttle-box sessions (S-C vs. NS-C: $p < 0.05$, days, 3, 4, 5). Subchronic injections of morphine (4 mg/

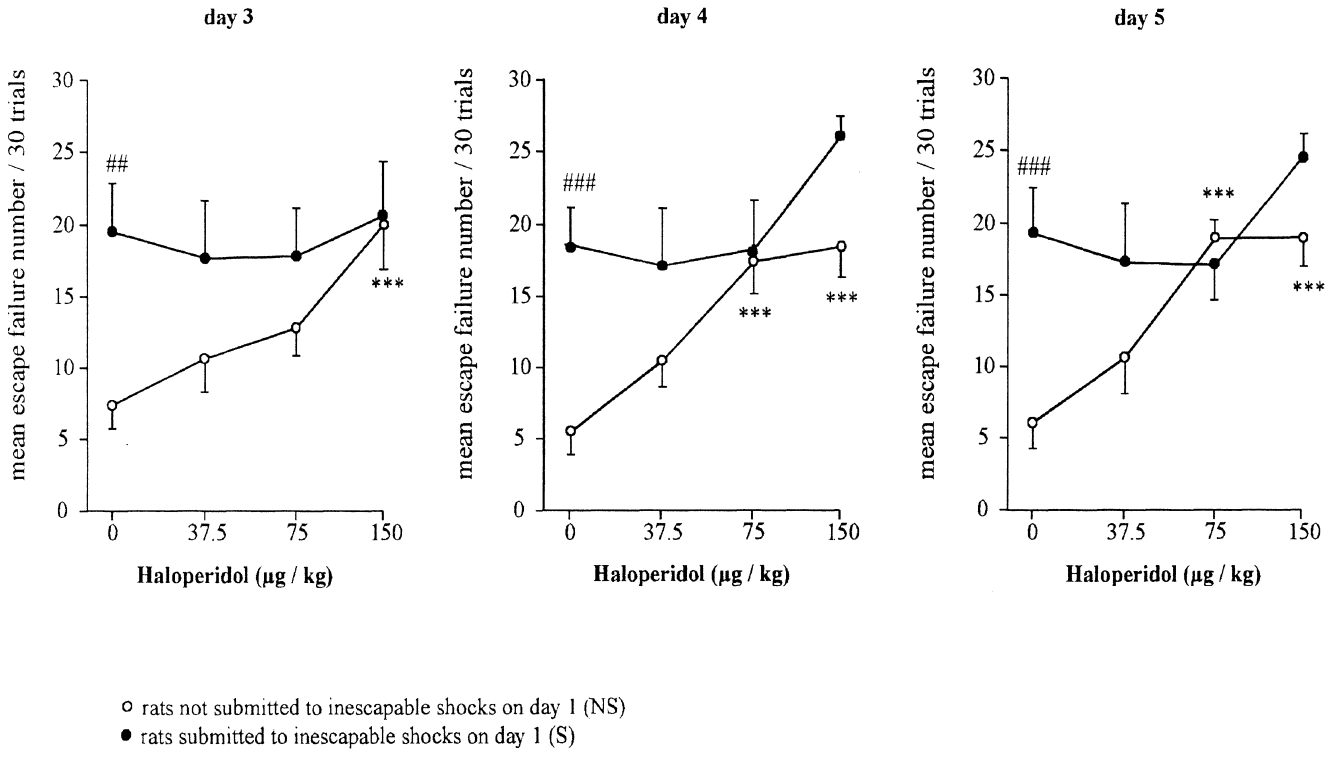

FIG. 1. Influence of haloperidol on escape behavior in the learned-helplessness paradigm. Data are the mean escape failure number (± SEM) during the 30 trials of the three daily shuttle-box sessions, on days, 3, 4, and 5, after exposure to inescapable shocks (S-groups) or not (NS-groups). Haloperidol (37.5 μg/kg; 75 μg/kg) was administered intraperitoneally 15 min before each shuttle-box session in both S-groups and NS-groups. #: Indicates that mean escape failure number of (S) control rats injected with saline statistically differs from (NS)-control rats (#$p < 0.05$, ##$p < 0.01$, ###$p < 0.001$). *:Indicates that the escape response of (NS) haloperidol-treated groups statistically differs from the (NS) control group (*$p < 0.05$; **$p < 0.01$, ***$p < 0.001$).

kg, a.m., and 2 mg/kg, p.m. for 5 days) reversed IS-induced escape deficit during all three shuttle-box sessions. Although nonstressed rats treated with this regimen of morphine performed better than their control counterparts (NS-C), this difference did not reach a statistical level of significance (Fig. 2A). Interestingly, an acute dose of haloperidol (37.5 μg/kg), although ineffective by itself, significantly and completely antagonized the response to morphine in previously stressed rats (Fig. 2A). However, haloperidol did not impair the escape response of nonstressed rats treated with morphine (Fig. 2A).

Intertrial crossings (ITC) (Fig. 2B). The Kruskal–Wallis ANOVA on ranks indicated out that a subchronic administration of morphine (6 mg/kg/day × 5 days) induced a significantly increased number of ITCs in both NS and S groups. This effect of morphine on ITC was enhanced on reexposure to the shuttle-box test in both NS-M and S-M groups. At the dose used in the present experiments, haloperidol did not alter intertrial activity in either NS or S rats. When an acute injection of haloperidol was given 15 min before the shuttle-box sessions in morphine-treated rats, the mean value of ITC de-

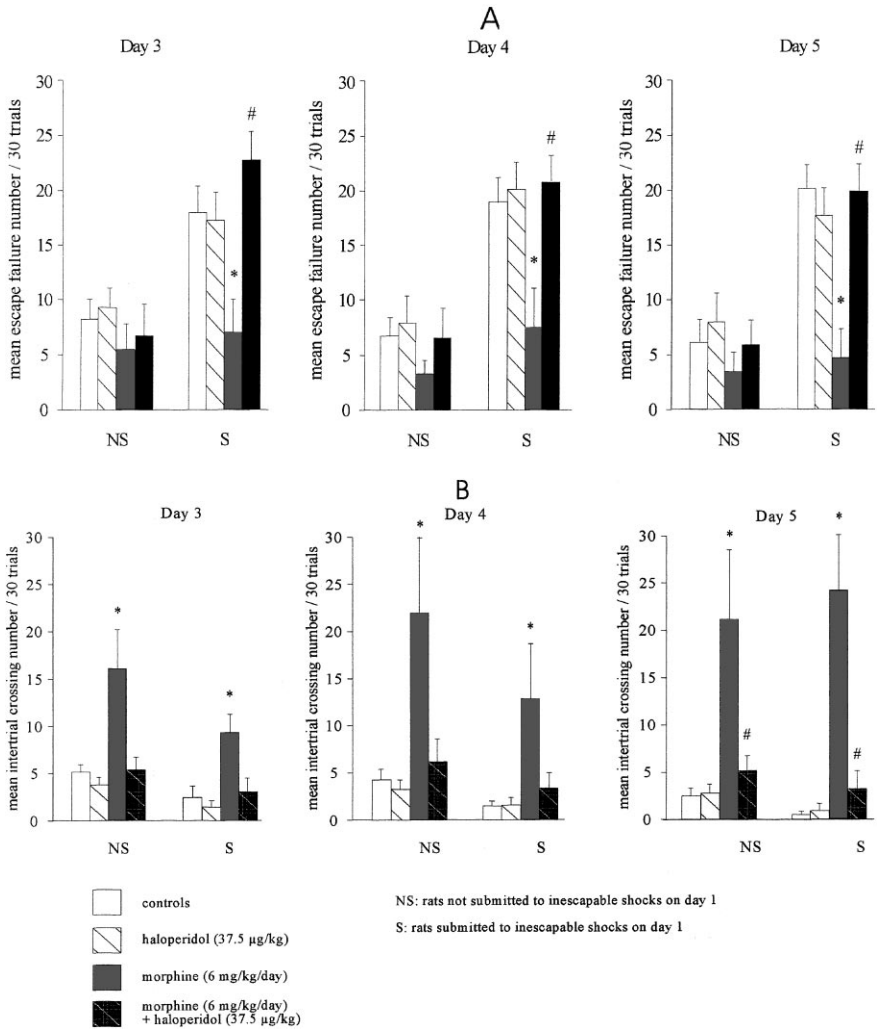

FIG. 2. Influence of haloperidol on escape response of morphine-treated rats (A) and on intertrial crossings (ITC) (B) in the learned-helplessness paradigm. Data are the mean escape failure number and ITC number (± SEM) during the 30 trials of the three shuttle-box sessions (days 3, 4, 5) after exposure to inescapable shocks (S) or not (NS). Both (NS) and (S) rats were treated with either saline (controls), morphine (6 mg/kg/day) or morphine (6 mg/kg/day)–haloperidol (37.5 μg/kg) combination. Morphine was injected subcutaneously during 5 consecutive days, the first injection taking place 4 h after inescapable shocks pretraining or restraint on day 1, and then twice daily (4 mg/kg in the morning and 2 mg/kg in the afternoon). For the morphine–haloperidol treatment, haloperidol (37.5 μg/kg) was injected intraperitoneally only in the morning on days 3, 4, and 5, 15 min after morphine and 15 min before the shuttle-box test. *:Indicates that (NS) rats treated with morphine alone or (S) rats treated with morphine alone statistically differ from their respective control groups ($*p < 0.05$). #:Indicates that (NS) rats treated with morphine–haloperidol combination statistically differ from the (NS) morphine-treated group or that (S) rats treated with morphine–haloperidol combination statistically differ from the (S) morphine-treated group ($\#p < 0.05$).

clined whatever the level of stress, and with a more pronounced inhibition of the effect of morphine during the last shuttle-box session (Fig. 2B). Despite the three- to eightfold drop in mean ITC values in NS-MH and S-MH groups compared with NS-M and S-M groups, respectively, during the 3 days of shuttle-box test these differences were statistically significant on the last day only.

Relationship between EF and ITC. Spearman's correlation on ranks indicated a significant negative correlation between these two parameters in S-M and S-MH rats (S-M: $r = -0.490$, $p = 0.015$; S-MH: $r = -0.394$, $p = 0.021$), with the number of EFs decreasing while the number of ITCs increased in these two groups. A negative correlation was also found for the NS-MH rats ($r = -0.488$, $p = 0.004$), but no significant correlation between the two parameters was found for the NS-M rats ($r = 0.016$, $p = 0.937$), this last group displaying no significant improvement in escape behavior compared with the NS-C group (Fig. 2A).

DISCUSSION

Several points emerged from the data reported here: (a) acute injections of haloperidol impaired the escape behavior of nonstressed rats dose dependently (Fig. 1). (b) In contrast, 6 mg/kg/day of morphine significantly improved the escape behavior of previously stressed rats (Fig. 2A), and significantly increased the number of ITCs in both NS and S rats (Fig. 2B). (c) A dose of haloperidol, although ineffective by itself (37.5 μg/kg, IP), completely suppressed morphine-induced reversal of EF in stressed rats (Fig. 2A), and altered morphine-induced increase in ITCs in both NS and S rats (Fig. 2B).

Few investigators have recorded intertrial activity (ITCs) during the shuttle-box test. However, Geoffroy and Christensen (16) have suggested that intertrial activity is an indication of "nonspecific motor activity" (or spontaneous motor activity) and they showed the usefulness of measuring it to assess a possible contribution of psychostimulant effects of drugs in the learned-helplessness paradigm. On this basis, we found it helpful to count ITCs in parallel with the EFs to detect whether a change in locomotor activity might participate in the behavioral interaction between morphine and haloperidol studied here.

Nonstressed rats receiving 75 or 150 μg/kg of haloperidol displayed a significant increase in EFs, so that their escape behavior became similar to stressed rats injected with vehicle. These results support previous reports indicating the induction of an escape deficit in nonstressed mice given 75 μg/kg of haloperidol 30 min before a shuttle-box test (4) and in nonstressed rats given 20 mg/kg of haloperidol 24 h before a shuttle-box test (40). Likewise, haloperidol (0.3 mg/kg, IP) increased the duration of immobility in the forced swimming test (FST) in mice (25). In this first series of experiments we did not report the effects of haloperidol on ITCs because of the low number of ITCs performed by rats in the NS-C group. A similar inhibition of mesolimbic DA activity may underly the behavioral effects of haloperidol and uncontrollable stress in the learned-helplessness paradigm as both of them have been shown to interact with this system [see (12), for review]. Haloperidol is largely considered as a preferential D_2-DA receptor antagonist. However, it binds to other types of receptors; in addition to its high D_2-DA receptor blocking potency, haloperidol is a putative sigma-1 ligand, displaying similar nanomolar affinity for both sites (28, 43, 45). Hence, both sigma receptor blockade and D_2-DA receptor blockade may be involved in the behavioral effect of haloperidol in the learned-helplessness model, mainly because both types of receptors were found in brain areas associated with the control of memory, emotion, and motor activity (13, 20, 35, 47). To elucidate the contribution of D_2-DA receptor site in this effect of haloperidol, we would need to study the effects of sulpiride, a selective D_2-DA receptor antagonist devoid of affinity for sigma sites (20, 38). In any case, changes in DA neurotransmission may be a final pathway to the effects of haloperidol because some experimental evidence supports the view that sigma receptors regulate dopaminergic function in nigrostriatal, mesocortical, and mesolimbic areas (21, 47). On the other hand, it is very unlikely that D_1-DA- and 5 HT_{2A} serotonergic-receptor subtypes were involved in this effect of haloperidol because Matsubara et al. (36) reported the absence of occupation of these sites with acute intraperitoneal doses of haloperidol up to 0.5 mg/kg in in vivo assays. In addition, in vitro receptor binding and in vivo receptor occupancy assays clearly demonstrated that haloperidol has about 20 and 14 times more affinity to D_2-DA receptors than 5 HT_{2A}-serotonergic receptors and α_1-adrenergic receptors, respectively, and its affinity for D_1- and D_3-DA receptor subtypes is 200 and 20 times lower than for D_2-DA sites, respectively (43).

Interestingly, naloxone, a selective opioid antagonist, displayed the same activity profile as haloperidol in our model (7), i.e., both drugs mimicked the behavioral deficit induced by an uncontrollable stress despite their divergent receptor binding affinities. Therefore, a dysregulation of both dopaminergic and opioid neurotransmission is likely to contribute to the expression of stress-induced escape deficit. Moreover, biochemical studies have indicated a regulation of dopamine neurotransmission by endogenous opioid release in response to stressful event [see the introductory paragraphs; (11, 22, 37)].

As shown in Fig 2, morphine (6 mg/kg/day, SC) suppressed the inescapable shock-induced escape deficit and stimulated intertrial activity in both nonstressed and stressed groups as previously seen (7). Morphine-induced reversal of escape failures was completely suppressed by haloperidol by the first shuttle-box session (Fig. 2A), whereas morphine-induced increased intertrial activity was significantly antagonized by haloperidol during the third shuttle-box session only (Fig. 2B). The lack of a significant antagonization of morphine-induced stimulation of intertrial activity during the first two shuttle-box sessions was probably due to the wide interindividual variability of ITC values within the different treated groups. Consequently, increasing the number of rats per group would probably produce a statistically significant difference. An interindividual variability of ITC values has previously been reported with morphine (7) and D_2-DA agonists (16). This behavioral antagonism cannot be ascribed to a direct competition at opioid receptor sites; haloperidol shows no affinity for mu, delta, and kappa opioid receptors, and a high affinity for the sigma receptor (43) for which morphine has negligible affinity (27). It is more likely that blockade of postsynaptic D_2-DA receptor sites by haloperidol prevents morphine indirectly enhancing postsynaptic DA neurotransmission, probably via the stimulation of mu-opioid receptors located on DA neuron terminals. These results indicate that both a mu-opioid mediation and a D_2-DA mediation are necessary for the expression of morphine activity in the learned-helplessness paradigm. However, a role for a sigma mediation in the behavioral interaction between morphine and haloperidol cannot be completely ruled out, as no information is available about a possible indirect functional interaction of morphine with sigma transmission. Additional studies with the selective D_2-DA receptor antagonist, sulpiride, would be helpful to de-

lineate the respective roles of DA and sigma mediation in the suppression of morphine effects by haloperidol in the learned-helplessness paradigm. Anyway, this study shows that opioid and DA neurotransmissions act concomitantly in the learned helplessness model that involves motivational, emotional, and motor components (19, 33, 50).

We emphasize the statistical correlation between the improvement of escape behavior and the increased intertrial activity in S-M rats. In addition, the suppression by haloperidol of morphine-induced reversal of EF in stressed rats was significantly correlated with the suppression of morphine-induced enhancement of intertrial activity. A reversal of escape deficit along with an increased intertrial activity was observed with various D_2-DA receptor agonists and was attributed to their psychostimulant properties, whereas well-known antidepressant drugs do not alter intertrial activity (16). Together, these data demonstrate that the behavioral interaction between morphine and haloperidol in the learned-helplessness paradigm results from the suppression of morphine-induced hyperlocomotor activity through a dopaminergic mechanism. Thus, the effects of morphine in the learned-helplessness paradigm may be related to its psychostimulant-like activity produced by an indirect DA stimulation rather than a pure antidepressant profile.

REFERENCES

1. Abercrombie, E. D.; Keefe, K. A.; Di Frischia, D. F.; Zigmond, M. J.: Differential effects of stress on in vivo dopamine release in striatum, nucleus accumbens and medial frontal cortex. J. Neurochem. 52:1655–1658; 1989.
2. Akil, H.; Watson, S. J.; Young, E.; Lewis, M.E.; Khachaturian, H.; Walker, J. M.: Endogenous opioids: Biology and function. Annu. Rev. Neurosci. 7:223–255; 1984.
3. American Psychiatry Association. DSM IV: Diagnostic and statistical manual of mental disorders, 4th ed. Washington, DC: American Psychiatric Press; 1994:320–327.
4. Anisman, H.; Ritch, M.; Sklar, L. S.: Noradrenergic and dopaminergic interactions in escape behavior: Analysis of uncontrollable stress effects. Psychopharmacology (Berlin) 74:263–268; 1981.
5. Anisman, H.; Zacharko, R. M.: Multiple neurochemical and behavioral consequences of stressors: Implications for depression. Pharmacol. Ther. 46:119–136; 1990.
6. Berger, A.; Nemeroff, C. B.: Opioid peptides in affective disorders. In: Meltzer, H. Y., eds. The third generation of progress, Psychopharmacology, chapt. 63. New York: Raven Press; 1987: 637–646.
7. Besson, A.; Privat, A. M.; Eschalier, A.; Fialip, J: Effects of morphine, naloxone and their interaction in the learned helplessness paradigm in rats. Psychopharmacology (Berlin) 123:71–78; 1996.
8. Borsini, F.; Lecci, A.; Macinell, A.; D'Aranno, V.; Meli, A: Stimulation of dopamine D2 but not D1 receptors reduces immobility of rats in the forced swimming test. Implication for antidepressant activity. Eur. J. Pharmacol. 148:301–307; 1988.
9. Briley, M.; Prost, J. F.; Moret, C.: Preclinical pharmacology of minalcipram. Intern. Clin. Psychopharmacol. 4:9–14; 1996.
10. Brown, A. S.; Gershon, S.: Dopamine and depression. J. Neural Transm. Gen. Sec. 91:75–109; 1993.
11. Cabib, S.; Oliverio, A.; Puglisi-Allegra, S.: Stress-induced decrease of 3-methoxytyramine in the nucleus accumbens of the mouse is prevented by naltrexone pretreatment. Life Sci. 45:1031–1037; 1989.
12. Cabib, S.; Puglisi-Allegra, S.: Stress, depression and the mesolimbic dopamine system. Psychopharamcology (Berlin) 128:231–342; 1996.
13. Cooper, J. R.; Bloom, F. E.; Roth, R. H.: The biochemical basis of neuropharmacology, 6th ed. New York: Oxford University Press; 1991:285–337.
14. Di Chiara, G.; Alan North, R.: Neurobiology of opiate abuse. Trends Pharmacol. Sci. 13:185–192; 1992.
15. Emrich, H. M.: A possible role of opioid substances in depression. In: Costa, E.; Racagni, G., eds. Typical and atypical antidepressants: Clinical practice, advances in biochemical psychopharmacology, vol. 32. New York: Raven Press; 1982:77–83.
16. Geoffroy, M.; Christensen, A. V.: Psychomotor stimulants versus antidepressants in the learned helplessness model of depression. Drug Dev. Res. 29:48–55; 1993.
17. Gessa, G. L.: Dysthymia and depressive disorders—Dopamine hypothesis. Eur. Psychiatry 11 (Suppl. 3):S123–S127; 1996.
18. Gysling, K.; Wang, R. Y.: Morphine-induced activation of A10 dopamine neurons in the rat. Brain Res. 277:119–127; 1983.
19. Harris, J. C.: Experimental animal modeling of depression and anxiety. Psychiatr. Clin. North Am. 12:815–835; 1989.
20. Heroux, A. J.; Tam, S. W.; De Souza, E. B.: Autoradiographic identification and characterization of sigma receptors in guinea-pig brain using 1(cyclopropylmethyl)-4-(2′-4′′-fluorophenyl)-2′-oxo-ethyl) piperidine, [^{3}H]DuP734, a novel sigma ligand. Brain Res. 598:76–86; 1992.
21. Iyengar, S.; Dilworth, V. M.; Mick, S. J.; Contreras, P. C.; Monahan, L. B.; Rao, T. S.; Wood, P. L.: Sigma receptors modulate both A9 and A10 dopaminergic neurons in the rat brain: Functional interaction with NMDA receptors. Brain Res. 524:322–326; 1990.
22. Kalivas, P. W.; Abhold, R.: Enkephalin release into the ventral tegmental area in response to stress: Modulation of mesocorticolimbic dopamine. Brain Res. 44:339–348; 1987.
23. Katoh, A.; Nabeshima, T.; Kameyama, T.: Behavioral changes induced by stressful situations: Effects of enkaphalins, dynorphin, and their interaction. J. Pharmacol. Exp. Ther. 253:600–607; 1990.
24. Kendler, K. S.; Kessler, R. C.; Walters, E. E.; Mac Lean, C.; Neale, M. C.; Health, A. C.; Eaves, L. J.: Stressful life events, genetic liability, and onset of major depression in women. Am. J. Psychiatry 152:833–842; 1995.
25. Kitamura, Y.; Nagatami, T.: Buspirone enhances immobility in the forced swim test in mice. Pharmacol. Biochem. Behav. 55: 445–451; 1996.
26. Koob, G. F.: Drug of abuse: Anatomy, pharmacology and function of reward pathways. Trends Pharmacol. Sci. 13:177–184; 1992.
27. Largent, B. L.; Wilkstrom, H.; Gundlach, A. L.; Snyder, S. H.: Structural determinants of sigma receptor affinity. Mol. Pharmacol. 32:772–784; 1987.
28. Leysen, J. E.; Janssen, P. M.; Gonmeren, W.; Wynants, J.; Pauwels, P. J.; Janssen, P. A.: In vitro and in vivo receptor binding and effects on monoamine turnover in rat brain regions of the novel antipsychotics risperidone and ocaperidone. Mol. Pharmacol. 41:494–508; 1992.
29. Leyton, M.; Stewart, J.: Preexposure to footshock sensitizes the locomotor response to subsequent systemic morphine and intranucleus accumbens amphetamine. Pharmacol. Biochem. Behav. 37:303–310; 1990.
30. Llorens-Cortes, C.; Pollard, H.; Schwartz, J. C.: Localization of opiate receptors in substantia nigra: Evidence by lesion studies. Neurosci. Lett. 12:321–326; 1979.
31. Magnus-Ellenbroek, B.; Havemann Reinecke, U.: Morphine-induced hyperactivity in rats. A rebound effect. Naunyn Schmiedebergs Arch. Pharmaco. 347:635–642; 1993.
32. Maïer, S. F.; Seligman, M. E. P.: Learned helplessness: Theory and evidence. J. Exp. Psychol. Gen. 1:3–46; 1976.
33. Martin, P.; Soubrie, P.; Simon, P.: Noradrenergic and opioid mediation of tricyclic-induced reversal of escape deficits caused by inescapable shocks pretreatment in rats. Psychopharmacology (Berlin) 90:90–94; 1986.
34. Martin, P.; Guillou, N.; Lacroix, P.; Billardon, M.: Effects of co-administration of anxiolytic and antidepressant drugs in the learned helplessness paradigm: Impact of hydroxyzine. Encephale 22:270–279; 1996.
35. Mash, D. C.; Zabetian, C. P.: Sigma receptors are associated with

cortical limbic areas in the primate brain. Synapse 12:195–205; 1992.
36. Mastsubara, S.; Matsubara, R.; Kusumi, I.; Koyama, T.; Yamashita, I.: Dopamine D_1, D_2 and serotonin-2 receptor occupation by typical and atypical antipsychotic drugs in vivo. J. Pharmacol. Exp. Ther. 265:498–508; 1993.
37. Miller, J. D.; Speciale, S. G.; McMillen, B. A.; German, D. C.: Nalaxone antagonism of stress-induced augmentation of frontal cortex dopamine metabolism. Eur. J. Pharmacol. 98:437–439; 1984.
38. Okuyama, S.; Chaki, S.; Yae, T.; Nakazato, A.; Muramatsu, M.: Autoradiographic characterization of binding sites for [^{3}H]NE-100 in guinea pig brain. Life Sci. 57:33–337; 1995.
39. Overmier, J. B.; Seligman, M. P.: Effects of inescapable shocks upon subsequent escape and avoidance learning. J. Comp. Physiol. Psychol. 63:23–33; 1967.
40. Petty, F.; Kramer, G.; Moeller, M.: Does learned helplessness induction by haloperidol involve serotonin mediation. Pharmacol. Biochem. Behav. 48:671–676; 1994.
41. Raevskii, K. S.; Sotnikova, T. D.; Gainetdinov, R. R.: The brain dopaminergic systems: Receptor heterogeneity, functional role and pharmacological regulation. Uspekhi Fiziologicheskikh Nauk. 27:3–29; 1996.
42. Rosetti, Z. L.; Lai, M.; Hmadan, Y.; Gessa, G. L.: Depletion of mesolimbic dopamine during behavioral despair: Partial reversal by chronic imipramine. Eur. J. Pharmacol. 243:313–315; 1993.
43. Schotte, A.; Janssen, P. F.; Gommeren, W.; Luyten, W. H.; Van Gompel, P.; Lesage, A. S.; De Loore, K.; Leysen, J. E.: Risperidone compared with new and reference antipsychotic drugs. In vitro and in vivo receptor binding. Psychopharmacology (Berlin) 124:57–73; 1996.
44. Shippenberg, T. S.; Bals-Kubik, R.; Herz, A.: Examination of the neurochemical substrates mediating the motivational effects of opioids: Role of the mesolimbic dopamine system and D1-vs D-2 dopamine receptors. J. Pharmacol. Exp. Ther. 265:53–59; 1992.
45. Su, T. P.; Schell, S. E.; Ford-Rice, F. Y.; London, E. D.: Correlation of inhibitory potencies of putative antagonists for sigma receptors in brain and spleen. Eur. J. Pharmacol. 148:467–470; 1988.
46. Rejedor-Real, P.; Mico, J. A.; Maldonado, R.; Roques, B. P.; Gibert-Rahola, J.: Effects of mixed (RB38A) and selective (RB38B) inhibitors of enkephalin degrading enzymes on a model of depression in the rat. Biol. Psychiatry 34:100–107; 1993.
47. Wheaterspoon, J. K.; Gonzalez-Alvear, G. M.; Frank, A. R.; Werling, L. L.: Regulation of [^{3}H] dopamine release from mesolimbic and mesocortical areas of guinea pig brain by sigma receptors. Schizophr. Res. 21:51–62; 1996.
48. Willner, P.: Dopamine and depression: A review of recent evidence. Brain Res. Rev. 6:211–246; 1983.
49. Willner, P.: Validation criteria for animal models of human disorders: Learned helplessness as a paradigm case. Prog. Neuropsychopharmacol. Biol. Psychiatry 10:677–690; 1986.
50. Willner, P.: Animal model as simulation of depression. Trends Neurosci. 121:131–136; 1991.
51. Zacharko, R. M.; Anisman, H.: Stressor-induced anhedonia and the mesolimbic dopamine system. Neurosci. Biobehav. Rev. 15: 391–405; 1991.
52. Zacharko, R. M.; Bowers, W. J.; Kokkinidis, L.; Anisman, H.: Region specific reductions of intracranial self-stimulation after incontrollable stress: possible effects on reward processes. Brain Res. 9:129–141; 1983.