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Effects of Neonatal Naltrindole Treatment on Antinociceptive and Behavioral Responses to μ and κ Agonists in Rats

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FERNÁNDEZ, B., M. T. ANTELO, I. KITCHEN AND M. P. VIVEROS. *Effects of neonatal naltrindole treatment on antinociceptive and behavioral responses to μ and κ agonists in rats.* PHARMACOL BIOCHEM BEHAV **62**(1) 145–149, 1999.—The effects of a daily injection of the δ selective opioid antagonist naltrindole (1 mg/kg), from birth to postnatal day 19, on antinociceptive and behavioral responses to the μ selective agonist alfentanil (65 μg/kg) and the κ selective agonist CI-977 (50 μg/kg) in 20-day-old male rats were investigated. Antinociception was assessed using the tail immersion test and behavioral testing was performed by employing an open field. The functional blockade of the δ receptor by naltrindole blocked the antinociceptive response to alfentanil but did not affect the antinociception induced by CI-977. The effects of alfentanil (increased exploration) and CI-977 (a marked hypoactivity) in the open field were not modified by neonatal naltrindole treatment. The results suggest a functional interaction between δ and μ receptors in the postnatal period but not between δ and κ receptors. The data also suggest differences in the δ and μ receptors interacting in the modulation of antinociception and those involved in behavioral responses in the open field. © 1998 Elsevier Science Inc.

Neonatal rats Naltrindole Alfentanil CI-977 Antinociception Open field

THERE is substantial evidence indicating the existence of critical periods during early postnatal development of the endogenous opioid system in the rat, in which the system is particularly sensitive to environmental insults and pharmacological treatments with opioid compounds (1,3,4,10,24,25,30). Some of these studies have shown that chronic treatments with opioid agonists or antagonists produce downregulation (24,30) or upregulation (1,10,25) in the opioid receptors, pointing to the plasticity of the opioid system. Many of these studies have used compounds that are not selective for the opioid receptor subtypes, such as morphine or the general antagonists naloxone and naltrexone. There is also much information about the existence of interactions and cooperativity between μ and δ receptors in adult rodents. These kind of interactions, which affect antinociceptive responses (8,20,21), the development of physical dependence on morphine (28), and brain-stimulation reward (6), may have implications for the therapeutic use of opioids. However, there is little evidence about this functional μ–δ-receptor interactions in newborn rats. Functional antinociceptive responses to μ-agonists, as well as μ opioid tolerance and dependence, have been demonstrated in neonatal rats (12,18,32). For the κ-opioid receptor, antinociceptive and behavioral responses to κ agonists have revealed striking differences between adult and neonatal rats (13,14,15,19). Information on δ-mediated responses in development is scarcer, in part due to the lack of availability of high-affinity agonists (7) and antagonists (22,29) until recently. When responses to δ-agonists have been reported (2,13,15), these show a lack of functional responses in preweanling animals possibly associated with the late development of this receptor (16,18).

To address the question of whether functional interactions exist between δ and μ receptors or δ and κ receptors in the neonatal period, we have studied the effects of functional blockade of neonatal σ receptors by a chronic treatment with naltrindole and measured antinociceptive and behavioral re-

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sponses to the selective μ agonist alfentanil and to the κ selective agonist CI-977 in neonatal rats.

METHOD

Animals and Neonatal Treatments

Experiments were performed on Wistar albino male rats from the animal house of the Universidad Complutense of Madrid, which is served by Harlan Interfauna Ibérica S.A. (Barcelona, Spain). The animals were maintained at a constant temperature of 20°C and in a reverse 12-D:12 L cycle (lights on at 2000 h), with free access to food (commercial diet for rodents A04/A03; Panlab, Barcelona, Spain) and water. Male rats were mated with females (one male × two females) and sperm-positive females were then rehoused in individual cages for the duration of pregnancy. On the day of birth (postnatal day 0), litters were sex balanced and culled to 10 ± 1 pups per dam. Although only the males were used in the behavioral tests, the whole litters remained with their mothers to avoid interferences due to sexual isolation (3,27). The pups were marked subcutaneously with ink to identify each of them. From the day of birth to day 19 half of the animals within each litter (males and females) received a daily SC injection of naltrindole (RBI) (1 mg/kg, 1 ml/kg) and the other half a SC injection with the same volume of 0.9% saline solution. Taken into account this neonatal treatment plus the different acute treatments detailed below, each experimental group contained individuals from a minimum of four different litters, which were tested on at least two different days to minimize interlitter and interday variability. All experimental procedures were carried out between 0930 and 1430 h. On the day of testing the animals were equilibrated in a quiet laboratory at least 1 h before experimental procedures were begun. Behavioral tests were carried out under the same illumination conditions as those in the animal facilities (red light).

Antinociceptive and Behavioral Responses to Alfentanil and CI-977

At 20 days of age, and within each of the above-mentioned neonatally treated groups, the following five acute treatment groups were studied: a control group, which received a single injection of 0.9% saline solution IP; two groups receiving either a single injection of alfentanil (Janssen) (65 μg/kg, IP) or CI-977 (Parke Davis) (50 μg/kg, IP) and two additional groups for antagonism studies that received an injection of naloxone (1 mg/kg, IP.) (RBI) before the administration of either alfentanil or CI-977. In previous studies (2,15) and from dose–response curves carried out in our laboratory (unpublished data) we found that the doses chosen for the μ- and κ-agonists produce submaximal responses in neonatal rats. The maximum dose volume was of 0.13 ml/20 g. Nociception was assessed using the tail immersion test with water at 50°C (17). Nociceptive responses (tail immersion latencies) were measured as the time elapsed prior to removal of the tail from the water surface, and a maximum 10-s cutoff was used. Response latencies were measured 15 min before administration of saline, alfentanil, or CI-977 and 5 min after treatment, a time point that represents peak antinociception for both alfentanil (2) and CI-977 (15) in this paradigm. The antagonist naloxone was administered 10 min before agonist administration. To compare the different treatments, antinociception was quantified as previously described (23) using the following formula:

$$\text{Latency quotient} = \frac{\text{response latency after treatment}}{\text{response latency before treatment}}$$

Behavioral testing was performed by using a square open field (60 × 60 × 45 cm) with a floor divided into 36 squares (10 × 10 cm). The duration of the test was 5 min. The parameters recorded were: external ambulation (number of entries to the external sectors situated in the periphery, by the walls) and internal ambulation (number of entries to the internal sectors situated in the central area), total ambulation (external + internal), frequency of rearing and facial grooming, and defecation score (number of boluses). The animals were tested individually in the open field, 5 min after the completion of the tail immersion test.

Statistical Analysis

Data from tail immersion (latency quotients) and open field were analyzed by two-way analysis of variance (ANOVA) (factors: neonatal treatment and acute treatment). The groups included in the analysis were chronically treated with saline or naltrindole, and subsequently studied for the acute effects of μ or κ agonists and antagonism by naloxone. Student–Newman–Keuls test with a level of significance set at $p < 0.05$ was used for posthoc comparisons. An unpaired Student *t*-test was employed to analyze basal nociceptive latencies.

RESULTS

Baseline Nociceptive Latencies

In order to evaluate the effect of neonatal naltrindole treatment on baseline nociceptive latencies (s) in the tail immersion, we analyzed the pretest data obtained from all the animals included in the study. There was no significant effect of neonatal treatment on the baseline responses (animals treated neonatally with saline: 1.71 ± 0.06, $n = 51$, animals treated neonatally with naltrindole: 1.89 ± 0.08, $n = 51$; $t(100) = -1.79$, $p = 0.08$).

Antinociceptive and Behavioral Responses to Alfentanil and CI-977

The analysis of the data from the tail immersion test revealed the following results: with respect to the μ agonist alfentanil, significant effects of the acute treatment, $F(2, 48) = 7.77$, $p = 0.001$, and a significant interaction between factors, $F(2, 48) = 4.88$, $p = 0.01$, were found, while the effect of the neonatal treatment was not significant. Posthoc comparisons showed that alfentanil caused a significant antinociception in the animals treated neonatally with saline, and that this effect was antagonized by naloxone. However, the μ-agonist did not induce any antinociceptive response in the animals treated neonatally with naltrindole (Fig. 1a). For the κ agonist CI-977, ANOVA revealed a significant effect of the acute treatment, $F(2, 63) = 10.72$, $p < 0.0001$, while the neonatal treatment and the interaction between treatments were not significant. CI-977 caused an antinociceptive effect in all the animals (treated neonatally with saline or naltrindole) that was antagonized by naloxone (Fig. 1b).

The analysis of the data from the open field did not reveal any significant interaction between neonatal and acute treatments for any of the parameters recorded, indicating that neonatal naltrindole treatment did not significantly modify the responses to alfentanil or CI-977 in this test. The analysis on the responses to alfentanil revealed significant effects of acute treatment for rearing behavior, $F(2, 49) = 16.81$, $p < 0.0001$, and a marginally significant effect for internal ambulation, $F(2, 49) = 3.01$, $p = 0.06$. In fact, the animals treated with alfentanil showed a significant increase in rearing behavior,

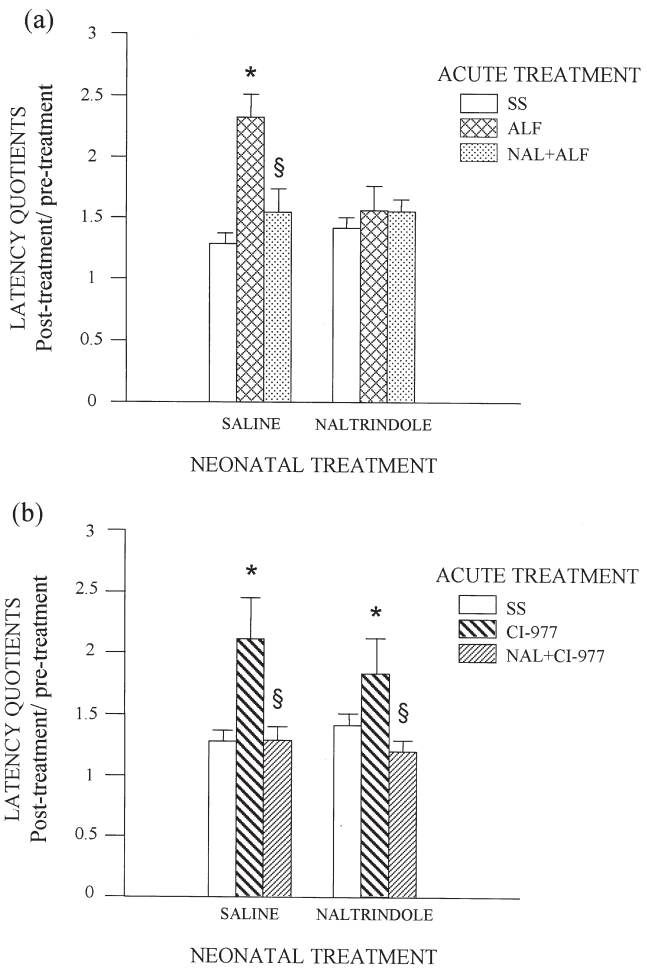

FIG. 1. Effects of neonatal naltrindole treatment on antinociceptive responses to alfentanil and CI-977 in the tail immersion test in 20-day-old male rats. The animals were treated neonatally with saline (SS) or naltrindole (NTI) (from birth to day 19) and subsequently studied for the acute effects of alfentanil (ALF) (a) or CI-977 (b) and antagonism by a previous injection with naloxone (NAL). (a) Histograms represent the mean ± SEM of 7–11 animals. Two-way ANOVA: significant effects of acute treatment and interaction between acute and neonatal treatments. Student–Newman–Keuls: $*p < 0.05$ vs. the control group treated acutely with SS, $\S p < 0.05$ vs. the group treated acutely with ALF. (b) Histograms represent the mean ± SEM of 10–15 animals. Two-way ANOVA: significant effect of acute treatment, no significant interaction between acute and neonatal treatments. Student–Newman–Keuls: $*p < 0.05$ vs. the control groups treated acutely with SS, $\S p < 0.05$ vs. the groups treated acutely with CI-977.

which was antagonized by naloxone, and a trend towards increased internal ambulation (Table 1). Analysis of responses to CI-977 showed significant effects of acute treatment for external ambulation, $F(2, 64) = 4.76$, $p = 0.01$, total ambulation, $F(2, 64) = 5.50$, $p < 0.01$, rearing behavior, $F(2, 64) = 85.58$, $p < 0.0001$, and grooming, $F(2,64) = 21.19$, $p < 0.0001$. Acute CI-977 treatment produced significant decreases in all these parameters. After the administration of the κ agonist sedation and straub tail responses were observed in some animals. Naloxone antagonized the effects of CI-977 on ambulation and reduced the effects on rearing, but did not prevent the effect on grooming (Table 1).

DISCUSSION

There is substantial evidence indicating the existence of interaction between μ and δ receptors in adults, and it has been hypothesized that μ and δ receptors may coexist in an opioid–receptor complex (8,20,21). Our results indicate that the functional blockade of the δ-receptors during postnatal days 0–19 blocked the antinociceptive response to the selective μ agonist alfentanil in preweanling rats, which suggest that this kind of interaction might exist during the period in which both receptors are in rapid development. These results also support previous studies suggesting that endogenous and exogenous σ agonists may enhance antinociceptive processes mediated by μ receptors (8,20,21). Thus, the chronic functional blockade of σ receptors by naltrindole during the postnatal period might have prevented a possible modulatory effect of endogenous σ agonists on μ receptors that might account for the inhibition of the antinociceptive action of alfentanil.

Although many studies have shown that κ-agonists in adults are relatively insensitive in thermal tests (9,19,26), this is not borne out in postnatal animals. The κ agonists PD 117302, U69593, and CI-977 produce a marked antinociception in the tail immersion test, in 5- and 10-day-old rats (15), U50,488 causes antinociception at day 10 (14), and CI-977 is effective in the tail-flick test in 3-day-old pups (19). Our results confirm that CI-977, at a dose of 50 μg/kg, was effective in producing antinociception at 20 days of age. As a whole, these data suggest that κ-mediated antinociception against thermal stimuli might be particularly important in the neonates in comparison with adult animals and this, indeed, might be biologically protective during early development. In contrast to the results obtained with alfentanil, the antinociception induced by the κ agonist was not significantly reduced by the neonatal treatment with naltrindole. This suggests that there is an interaction between μ and δ receptors, but not between κ and δ receptors in preweanling rats in relation to nociception.

Our previous studies have shown that μ agonists (morphine and DAGO) induced behavioral depression in neonatal rats of 5–20 days of age when tested in a cage similar to their usual home cage, i.e., under a low-stress condition (13). In contrast, the results of this work indicate that the selective μ agonist alfentanil caused increases in internal ambulation and rearing behavior in the open field, a test that involves a moderate degree of stress. These results suggest that alfentanil increased the exploratory activity, and this accords with previous studies showing that opioid substances may facilitate the exploratory behavior in novel or aversive environments (31). In contrast to the findings regarding the interaction between the neonatal naltrindole treatment and alfentanil-mediated antinociceptive responses, the open-field responses to alfentanil were not affected by neonatal treatment with the δ antagonist. This suggests that the μ and δ receptors interacting in the modulation of antinociception might be at least in part different from those involved in these type of behavioral responses in the open field.

In studies where behavioral responses of neonatal rats to κ-agonists have been investigated, these compounds induce hyperactivity in rats of 3, 5, 10, and 15 days of age (13,14,19). In contrast, κ-agonists, and in particular CI-977, typically induce hypoactivity in mature rats and mice (5,9,11,19). Our results indicate that CI-977 induced a marked hypoactivity in the open field at 20 days of age, which was reflected as significant decreases in external and total ambulation and rearing behavior, with this latter parameter being decreased by approximately 90%, with also significantly reduced grooming. These effects were not affected by the neonatal treatment

TABLE 1

EFFECTS OF ALFENTANIL AND CI-977 ON OPEN-FIELD ACTIVITY IN MALE RATS TREATED NEONATALLY WITH NALTRINDOLE AT 20 DAYS OF AGE

Treatments		Parameters					
Acute	Neonatal	EA	IA	TA	RB	G	DEF
SS	SS	169.6 ± 11.9	66.2 ± 6.9	235.8 ± 14.1	43.5 ± 3.0	3.4 ± 0.7	0.3 ± 0.2
	NTI	188.2 ± 15.8	59.8 ± 6.6	248.4 ± 19.2	40.3 ± 4.6	2.1 ± 0.4	0.1 ± 0.1
ALF	SS	169.9 ± 11.9	81.5 ± 7.2	251.4 ± 15.9	60.1 ± 5.3 *	2.1 ± 0.4	0.1 ± 0.1
	NTI	168.3 ± 9.5	79.7 ± 10.4	248.0 ± 13.2	56.6 ± 4.2	1.6 ± 0.4	0.1 ± 0.1
NAL + ALF	SS	144.1 ± 13.3	55.1 ± 10.7	199.3 ± 20.6	34.4 ± 3.2 §	3.0 ± 0.6	0.4 ± 0.3
	NTI	172.3 ± 15.6	72.6 ± 8.3	244.9 ± 21.2	33.1 ± 3.3	2.7 ± 0.8	0.6 ± 0.4
CI-977	SS	125.8 ± 18.4 *	42.3 ± 8.6	168.0 ± 25.5 *	7.7 ± 3.1 *	0.4 ± 0.3 *	0.2 ± 0.2
	NTI	134.5 ± 17.2	53.5 ± 6.8	188.0 ± 20.6	4.0 ± 1.2	0.0 ± 0.0	0.0 ± 0.0
NAL + CI977	SS	152.1 ± 12.5 ¶	61.4 ± 10.6	217.3 ± 20.4 ¶	21.6 ± 2.5 *¶	0.9 ± 0.3 *	0.6 ± 0.2
	NTI	198.8 ± 29.9	59.9 ± 4.9	258.7 ± 31.5	21.3 ± 2.4	1.1 ± 0.5	0.3 ± 0.2

Values are the mean ± SEM of 7–15 animals. SS: saline, NTI: naltrindole, ALF: alfentanil, NAL: naloxone, EA: external ambulation, IA: internal ambulation, TA: total ambulation, RB: rearing behaviour, G: grooming, DEF: defecation. Student–Newman–Keuls test: *$p < 0.05$ vs. the groups treated acutely with SS, § $p < 0.05$ vs. the groups treated acutely with ALF, ¶$p < 0.05$ vs. the groups treated acutely with CI-977.

with naltrindole. In a previous study (13) it was found that U50,488H supressed grooming and gnawing behavior in 20-day-old rats, while locomotor activity was relatively unaffected. Other data indicate that the effect of CI-977 on activity in 21-day-old animals appeared to be biphasic, with some stimulatory effects appearing at low doses (10 μg/kg) and a significantly decreased open field activity with 1000 μg/kg (19). Although this dose is much higher than that used in this study, it appears that at postnatal days 20–21 there are clear changes in the effects of CI-977 on the animals' activity when compared with those induced in younger rats. It has been proposed that this fact might reflect a developmental transition in the substrate subserving the effects of CI-977 on activity (19). Given the important role that the process of weaning appears to play in the development of δ-opioid receptors (16), the possible existence of an additional influence of weaning on the development of κ-mediated behavioral responses deserves further investigation.

In conclusion, the functional blockade of the δ-receptor by naltrindole blocked the antinociceptive response to alfentanil but did not affect the antinociception induced by CI-977, in preweanling rats. The results suggest a functional interaction between δ and μ receptors in the postnatal period but not between δ and κ receptors. To address the molecular basis of these functional responses, we are currently evaluating by autoradiographic mapping if and how the chronic administration of naltrindole influences the expression of μ, δ, and κ-opioid receptors.

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REFERENCES

1. Bardo, M. T.; Bhatnagar, R. K.; Gebhart, G. F.: Age-related differences in the effect of chronic administration of naloxone on opiate binding in rat brain. Neuropharmacology 22:453–461; 1983.
2. Crook, T. J.; Kitchen, I.; Hill, R. G.: Effects of the δ-opioid receptor antagonist naltrindole on antinociceptive responses to selective δ-agonists in post-weanling rats. Br. J. Pharmacol. 107:573–576; 1992.
3. De Cabo, C.; Kelly, M.; Kitchen I.; Viveros, P.: Effects of neonatal sexual isolation on μ receptor development and nociceptive responses in the rats. Neurosci. Res. Commun. 10:79–86; 1992.
4. De Cabo, C.; Kitchen, I.; Kelly, M.; Viveros, M. P.: Effects of β-funaltrexamine treatment and sexual isolation in the perinatal period on the development of μ-opioid receptors and nociception. Psychoneuroendocrinology 18:415–424; 1993.
5. DiChiara, G.; Imperato, A.: Opposite effects of mu and kappa opiate agonists on dopamine release in the nucleus accumbens and in the dorsal caudate of freely moving rats. J. Pharmacol. Exp. Ther. 244:1067–1080; 1988.
6. Duvauchelle, C. L.; Fleming, S. M.; Kornetsky, C.: Involvement of delta- and mu-opioid receptors in the potentiation of brain-stimulation reward. Eur. J. Pharmacol. 316:137–143; 1996.
7. Erspamer, V.; Melchiorri, P.; Falonieri-Erspamer, G.; Negri, L.; Corsi, R.; Severini, C.; Barra, D.; Simmaco, M.; Kreil, G.: Deltorphins: A family of naturally occurring peptides with high affinity and selectivity for delta-opioid binding sites. Proc. Natl. Acad. Sci. USA 86:5188–5192; 1989.
8. Heyman, J. S.; Vaught, J. L.; Mosberg, H. I.; Haaseth, R. C.; Porreca, F.: Modulation of μ-mediated antinociception by δ agonists in the mouse: Selective potentiation of morphine and normor-

phine by [D-Pen^2 ,D-Pen^5] enkephalin. Eur. J. Pharmacol. 165:1–10; 1989.
9. Hunter, J. C.; Leighton, G. E.; Meecham, K. G.; Boyle, S. J.; Horwell, D. C.; Rees D. C.; Hughes, J.: CI-977, a novel and selective agonist for the κ opioid receptor. Br. J. Pharmacol. 101:183–189; 1990.
10. Imai, Y.; Wang, J. B.; Moriwaki, A.; Uhl, G. R.: Opiate receptor: Developmental profile and regulation. Soc. Neurosci. Abstr. 20: 1727; 1994.
11. Jackson, A.; Cooper, S. J.: An observational analysis of the effect of the selective kappa opioid agonist, U50,488H, on feeding and related behaviours in the rat. Psychopharmacology (Berlin) 90: 217–221; 1986.
12. Jackson, H. C.; Kitchen, I.: Antinociceptive effects of the μ-selective agonist [D-Ala^2-MePhe^4-Glyol^5] enkephalin in 10-day old neonatal rats. Adv. Biosci. 75:487–490; 1989.
13. Jackson, H. C.; Kitchen, I.: Behavioural effects of selective μ, κ and δ opioid agonists in neonatal rats. Psychopharmacology (Berlin) 97:404–409; 1989.
14. Kehoe, P.; Boylan, C. B.: Behavioral effects of kappa-opioid-receptor stimulation on neonatal rats. Behav. Neurosci. 108:418–423; 1994.
15. Kitchen, I.; Kelly, M. D. W.; De Cabo, C.: κ- and δ-opioid receptor mediated antinociception in preweanling rats. Analgesia 1:512–515; 1995.
16. Kitchen, I.; Leslie, F. M.; Kelly, M.; Barnes, R.; Crook, T. J.; Hill, R. G., Bordosi, A.; Toth, G.; Melchiorri, P.; Negri, L.: Development of delta-opioid receptor subtypes and the regulatory role of weaning: Radioligand binding, autoradiography and *in situ* hybridization studies. J. Pharmacol. Exp. Ther. 275:1597–1607; 1995.
17. Kitchen, I.; McDowell, J.; Winder, C.; Wilson, J. M.: Low level lead exposure alters morphine antinociception in neonatal rats. Toxicol. Lett. 22:119–123; 1984.
18. McDowell, J.; Kitchen, I.: Development of opioid systems: Peptides, receptors and pharmacology. Brain Res. Rev. 12:397–421; 1987.
19. McLaughlin, C. R.; Tao, Q.; Abood, M. E.: Analysis of the antinociceptive actions of the κ-opiod agonist enadoline (CI-977) in neonatal and adult rats: Comparison to κ-opioid receptor mRNA ontogeny. Drug Alcohol Depend. 38:261–269; 1995.
20. Negri, L.; Improta, G.; Lattanzi, R.; Potenza, R. L.; Luchetti, F.; Melchiorri, P.: Interaction between the mu-agonist dermorphin and the delta-agonist [D-Ala^2, Glu^4] deltorphin in supraspinal antinociception and delta-opioid receptor binding. Br. J. Pharmacol. 116:2931–2938; 1995.
21. Porreca, F.; Heyman, J. S.; Mosberg, H. I.; Omnaas, J. R.; Vaught, J. L.: Role of μ and δ receptors in the supraspinal and spinal analgesic effects of [D-Pen^2 ,D-Pen^5] Enkephalin in the mouse. J. Pharmacol. Exp. Ther. 241:393–400; 1987.
22. Portoghese, P. S.; Sultana, M.; Takemori, A. E.: Naltrindole, a highly selective and potent non-peptide δ opioid receptor antagonist. Eur. J. Pharmacol. 146:185–186; 1988.
23. Pujol, A.; De Cabo, C.; Martín, M. I.; Viveros, M. P.: A developmental study on stress-induced antinociception measured by the tail electric stimulation test. Pharmacol. Biochem. Behav. 46:373–376; 1993.
24. Rimanóczy, A.; Vathy, I.: Prenatal exposure to morphine alters brain μ opioid receptor characteristics in rats. Brain Res. 690:245–248; 1995.
25. Rothman, R. B.; Bykov, V.; Long, J. B.; Brady, L. S.; Jacobson, A. E.; Rice, K. C.; Holaday, J. W.: Chronic administration of morphine and naltrexone up-regulate μ-opoid binding sites labeled by [^{3}H][D-Ala^2,MePhe^4,Gly-ol^5]enkephalin: further evidence for two μ-binding sites. Eur. J. Pharmacol. 160:71–82; 1989.
26. Schmauss, C.: Spinal κ-opioid receptor-mediated antinociception is stimulus specific. Eur. J. Pharmacol. 137:197–205; 1987.
27. Spear, L. P.; File, S. E.: Methodological considerations in neurobehavioral teratology. Pharmacol. Biochem. Behav. 55:455–457; 1996.
28. Suzuki, T.; Tsuji, M.; Mori, T.; Misawa, M.; Nagase, H.: Effect of naltrindole on the development of physical dependence on morphine in mice: A behavioral and biochemical study. Life Sci. 57:PL247–PL252; 1995.
29. Takemori, A. E.; Portoghese, P. S.: Selective naltrexone-derived opioid receptor antagonists. Annu. Rev. Pharmacol. Toxicol. 32: 239–269; 1992.
30. Tempel, A.; Habas, J.-E.; Paredes, W.; Barr, G. A.: Morphine-induced downregulation of μ-opioid receptors in neonatal rat brain. Dev. Brain Res. 41:129–133; 1988.
31. Van Abeelen, J. H. F.: Genetic control of hippocampal cholinergic and dynorphinergic mechanisms regulating novelty-induced exploratory behavior in house mice. Experientia 47:839–845; 1989.
32. Windh, R. T.; Little, P. J.; Kuhn, C. M.: The ontogeny of mu opiate tolerance and dependence in the rat: Antinociceptive and biochemical studies. J. Pharmacol. Exp. Ther. 273:1361–1374; 1995.