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The antinociceptive activity of κ- but not δ-opioid receptor agonists is maintained in morphine-tolerant neuropathic rats

Gwénaëlle Catheline [a], Valérie Kayser [a,*], Juhana J. Idänpään-Heikkilä [a,b], Gisèle Guilbaud [a]

[a] *Unité de Recherches de Physiopharmacologie du Système Nerveux, I.N.S.E.R.M. U161, 2 rue d'Alésia, 75014 Paris, France*
[b] *The Institute of Biomedicine, Department of Pharmacology and Toxicology, University of Helsinki, Helsinki, Finland*

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Abstract

The antinociceptive effect of the preferential μ-opioid receptor agonist morphine (1 mg/kg i.v.), the δ-opioid receptor agonists, DTLET ([D-Thr^2,Leu^5]enkephalin-Thr) (3 and 6 mg/kg i.v.) and BUBUC ([D-Cys(StBu)^2,Leu^5]enkephalin-Thr(OtBu) (3 mg/kg i.v.), and the κ-opioid receptor agonist U-69,593 (trans-3,4-dichloro-*N*-methyl-*N*-[7-(1-pyrrolidinyl)cyclohexil]benzeneacetamide methanesulfonate) (0.25, 0.5 and 0.75 mg/kg i.v.) was evaluated in mononeuropathic (chronic constriction of the common sciatic nerve) rats. The rats were pretreated s.c. with 10 mg/kg of morphine, or saline, twice daily from day 12 to day 16 after the surgery. In morphine-pretreated rats, the antinociceptive effect of morphine on the vocalization threshold to paw pressure was greatly reduced, as compared to the saline-pretreated group. The antinociceptive effect of DTLET and BUBUC had also disappeared in the morphine-pretreated rats. By contrast, the potent antinociceptive effect of U-69,593 was not affected by the morphine pretreatment. Furthermore, the effect of U-69,593 was reversed by the specific κ-opioid receptor antagonist nor-binaltorphimine (1 and 2 mg/kg i.v.). These results suggest that in mononeuropathic rats, morphine pretreatment results in cross-tolerance to δ- but not to κ-opioid receptor agonists.

Keywords: Neuropathic pain; Mononeuropathic rat; Morphine; Tolerance; Opioid receptor agonist

1. Introduction

Pain resulting from lesions of the peripheral or central nervous system, often referred to as neuropathic pain, is difficult to treat. In particular, the question whether opioids exert an analgesic effect on this type of pain remains controversial (Arner and Meyerson, 1988; Kupers et al., 1991; Rowbotham et al., 1991; Jadad et al., 1992). The development of tolerance to the analgesic effects of opioids complicates their use in patients with persistent pain (Portenoy, 1994). Moreover, tolerance is difficult to separate from the confounding effects of disease progression and increasing psychological distress, as these also require an increase in opioid dosage (Collin et al., 1993).

We have demonstrated the antinociceptive effects of systemic morphine (Neil et al., 1990; Attal et al., 1991, Kayser et al., 1995b) and various opioid receptor agonists (Desmeules et al., 1993a) in the vocalization threshold to paw pressure test using rats with peripheral mononeuropathy, which is an experimental model of neuropathic pain (Bennett and Xie, 1988; Attal et al., 1990). We have previously studied the possible development of tolerance to the effects of morphine in mononeuropathic rats using relatively low doses of morphine (3 mg/kg s.c. twice daily for 4 days, Neil et al., 1990). However, the antinociceptive effect of morphine was not modified by the pretreatment. Therefore, in the present study, we have used a higher pretreatment dose of morphine (10 mg/kg s.c. twice daily during 4 days). Since one strategy utilized in the management of opioid analgesic tolerance involves switching the type of opioid used, the question of whether morphine-pretreated neuropathic rats were also tolerant to the antinociceptive effects of other opioid receptor agonists was also addressed. A preliminary report of these experiments has been previously reported (Kayser et al., 1995a).

2. Materials and methods

2.1. Animals

Male Sprague-Dawley rats (Charles River, France) 9–10 weeks old, weighing 175–200 g on arrival were used

* Corresponding author. Tel.: (33-1) 4589-3662; Fax: (33-1) 4588-1304.

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($n = 126$). The animals were housed in standard rodent cages (4–5 in a cage) and allowed to habituate to the colony room for 1 week before surgery. The rats had free access to standard laboratory pellets and tap water.

The experiments were carried out according to the Ethical Guidelines of the International Association for the Study of Pain (Committee for Research and Ethical Issues of the IASP, 1983). In particular, the number of animals was kept to a minimum. For this reason, vehicle controls were excluded since we have previously shown that acute administration of i.v. saline does not change vocalization thresholds to paw pressure (Desmeules et al., 1993a).

2.2. Surgical procedure

The unilateral peripheral mononeuropathy was produced on the right hind paw according to the method described in detail previously (Bennett and Xie, 1988; Attal et al., 1990). Under sodium pentobarbital anesthesia (50 mg/kg i.p.), four loose ligatures (5/0 chromic catgut) with 1 mm spacing were placed around the sciatic nerve, taking care not to interrupt the epineural circulation. To minimize the discomfort after the surgery, the rats were housed in large cages with sawdust bedding.

Eight non-operated rats served as controls. Sham-operated rats were not used since we have previously demonstrated (Attal et al., 1990; Desmeules et al., 1995) that the vocalization threshold to hind paw pressure remained steady without significant differences from pre-operative values in these animals. In addition, we have shown that the effects induced by successive doses of morphine (0.1–1 mg/kg i.v.) on the sham-operated paw were not significantly different from those observed in non-operated rats (Attal et al., 1991).

2.3. Morphine pretreatment

All rats were pretreated with morphine hydrochloride (10 mg/kg) s.c. or the equivalent volume of saline. The injections were given twice daily (at 9.30 a.m. and 5.30 p.m.) for 4 days, beginning on day 12 after the surgery in the neuropathic rats. In these neuropathic animals, the effect of the pretreatment was thus tested on day 16 after surgery. At this time, the abnormal pain behavior is at a stable maximum (Bennett and Xie, 1988; Attal et al., 1990). The antinociceptive effect of morphine or the specific opioid receptor agonists was assessed 17 h after the last pretreatment injection of saline or morphine.

2.4. Antinociceptive testing

Test sessions were begun at 9.30 a.m. They were carried out in a quiet room, remote from the colony room. Vocalization thresholds were determined by a previously described modification of the Randall and Selitto method, where a constantly increasing pressure is applied to the hind paw until the rat squeaks (Attal et al., 1990). This response represents a more integrated nociceptive behavior than the withdrawal of the paw (Kayser and Guilbaud, 1990) and is especially sensitive to opioid analgesic compounds particularly in this pain model (Attal et al., 1991; Desmeules et al., 1993a; Lee et al., 1994).

The Basile analgesimeter (Ugo Basile, Comerio, Italy, tip diameter of the stylus: 1 mm) was used. Each animal was carefully handled and wrapped in a towel so that only the limbs and head were free. The paw was placed under the stylus and the probe was applied to the dorsal part of the paw between the 3rd and the 4th metatarsus (the sciatic nerve territory). The withdrawal reflex, that usually occurs before vocalization, was prevented by gently holding the hind paw in position under the pusher until the rat vocalized.

For each rat, a pre-operative threshold (mean of two consecutive stable thresholds, expressed in grams) and a day 12 post-operative threshold (two determinations, just before the beginning of the morphine or saline pretreatment) were determined. On day 16 after the surgical procedure, the rats were randomly assigned to groups of five for a series of tests. The body weight and a control threshold (mean of two consecutive stable determinations) were determined for each rat. After injecting the drugs, the nociceptive pressure thresholds were measured every 10 min until they had returned to the baseline. The experimenter was unaware of the drugs and doses used.

2.5. Drugs

The following drugs were used: morphine hydrochloride (1 mg/kg, Meram, Paris, France), the δ-opioid receptor agonists DTLET ([D-Thr^2,Leu^5]enkephalin-Thr, 3 and 6 mg/kg, Neosystem, Strasbourg, France) and BUBUC ([D-Cys(StBu)^2,Leu^5]enkephalin-Thr(OtBu), 3 mg/kg, Bale Biochimie, Voisin le Bretonneux, France), the κ-opioid receptor agonist U-69,593 (trans-3,4-dichloro-*N*-methyl-*N*-[7-(1-pyrrolidinyl)cyclohexil]benzeneacetamide methanesulfonate, 0.25, 0.5 and 0.75 mg/kg, Sigma, Saint Quentin Fallavier, France) and the κ-opioid receptor antagonist nor-binaltorphimine dihydrochloride (1 and 2 mg/kg, Bioblock, Illkirch, France). All drugs were diluted in sterile physiological saline and administered i.v. into the lateral tail vein. These doses have previously been shown to relieve abnormal responses to paw pressure in neuropathic rats (Attal et al., 1991; Desmeules et al., 1993a).

2.6. Statistical procedures

Vocalization thresholds are given in grams (g). Data are expressed as means ± S.E.M. A paired *t*-test was used to compare the thresholds obtained before and after the surgery and to compare the drug effects with the control value. An unpaired *t*-test was used to compare the thresholds obtained from the saline- and the morphine-pretreated groups, before the acute injection. A factorial analysis of variance (ANOVA) plus Fisher's post least significant

Table 1
Evolution of the vocalization thresholds to paw pressure (in g) for both hindpaws on day 12 and on day 16 after the surgery

	Before surgery	Day 12	Day 16
Saline-pretreated $n = 53$			
Nerve injured paw	339 ± 9	243 ± 9 [a]	231 ± 6
Contralateral paw	345 ± 9	336 ± 12	330 ± 6
Morphine-pretreated $n = 65$			
Nerve injured paw	336 ± 12	240 ± 12 [a]	204 ± 6 [b]
Contralateral paw	345 ± 12	330 ± 15	300 ± 6 [b]

The neuropathic rats were pretreated with saline or morphine (10 mg/kg s.c. twice daily) from day 12 to day 16. [a] $P < 0.001$, paired t test, compared to the pre-operative value. [b] $P < 0.01$, unpaired t test, compared to the saline-pretreated group on day 16.

difference (PLSD) test were used to compare the areas under the curve (AUC) between the groups. Differences between group means were regarded as being statistically significant when P values were less than 0.05.

3. Results

3.1. The effects of the saline and morphine pretreatments on the vocalization thresholds

Before the beginning of the saline or morphine pretreatment, the evolution of the thresholds in neuropathic rats was roughly comparable to that of our previous studies (Attal et al., 1991; Desmeules et al., 1995). Before the nerve ligature, the vocalization thresholds (336 ± 5 g and 345 ± 5 g, $n = 118$) obtained from the right and left hind paws, respectively, did not differ significantly. As reported earlier (Attal et al., 1991), the mean threshold of the nerve-injured side was markedly decreased on day 12 after the surgery (Table 1).

On the day of testing (day 16 after the surgery), the mean threshold of the nerve-injured paw was further decreased (Table 1), which could be attributed to the evolution of the disease (Attal et al., 1990). Interestingly, the mean thresholds of both the nerve-injured and contralateral paws of the morphine-pretreated were significantly lower than the saline-pretreated rats (Table 1). Likewise, there was a significant decrease in the mean vocalization threshold of the morphine-pretreated non-operated rats (285 ± 4 g vs 337 ± 3 g, $P < 0.01$, paired t-test, $n = 4$ in each group). These decreases in the thresholds were thus due to the morphine pretreatment.

3.2. Signs of withdrawal in the morphine-pretreated rats.

On the day of testing, the neuropathic rats presented piloerection and diarrhea which are discrete signs of opioid withdrawal. An increased sensitivity to touch was also noted. No obvious signs of withdrawal were observed in

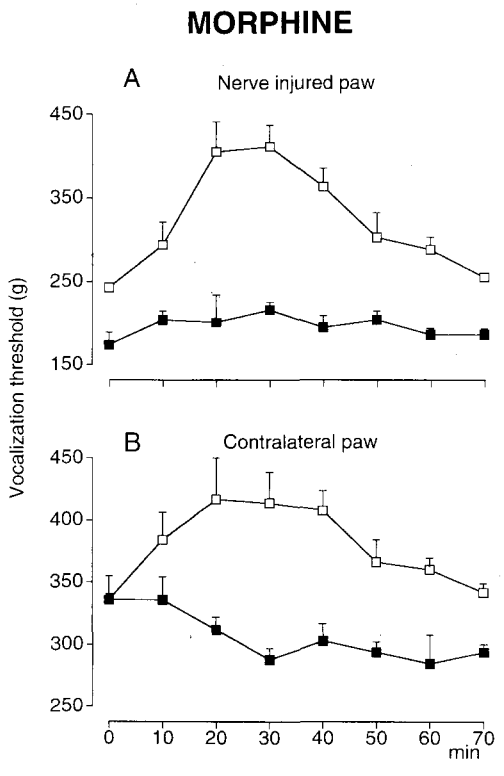

Fig. 1. The antinociceptive effect of morphine (1 mg/kg i.v.) on the vocalization threshold to paw pressure of the neuropathic rats pretreated with saline (□, $n = 8$) or morphine (■, $n = 6$). (A) nerve-injured paw, (B) contralateral paw.

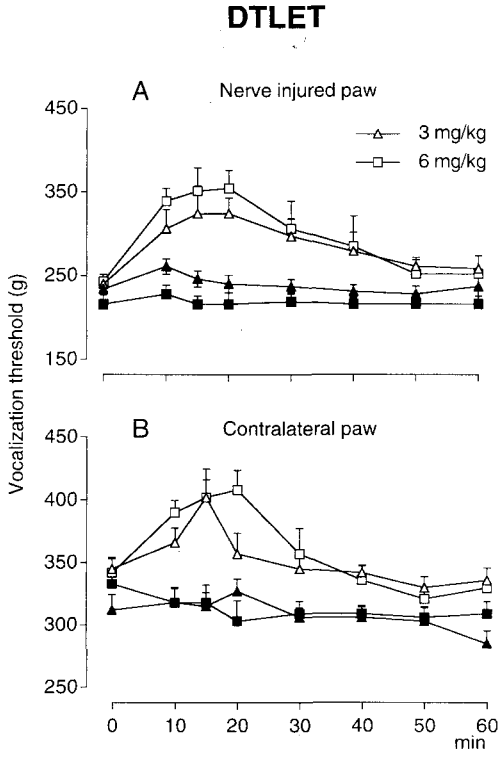

Fig. 2. The antinociceptive effect of DTLET (3 and 6 mg/kg i.v.) on the vocalization threshold to paw pressure of the neuropathic rats pretreated with saline (□, △, open symbols, 3 mg/kg $n = 7$ and 6 mg/kg $n = 5$) or morphine (■, ▲, 3 mg/kg $n = 10$ and 6 mg/kg $n = 6$). (A) nerve-injured paw, (B) contralateral paw.

the non-operated rats. Depending on the pretreatment, the body weights of the neuropathic rats were significantly different on the day of testing. The mean body weight (293 ± 16 g) of the saline-pretreated neuropathic rats (n = 53) was comparable to that of unpretreated neuropathic rats (Attal et al., 1991). The mean body weight (271 ± 15 g) of the morphine-pretreated neuropathic rats (n = 64) was significantly lower than that of the saline-pretreated neuropathic group ($P < 0.001$, unpaired t-test). The mean body weight (270 ± 4 g, n = 4) of the non-operated morphine-pretreated rats was significantly lower than that of the non-operated saline-pretreated animals (302 ± 6 g, n = 4, $P < 0.01$, unpaired t-test).

3.3. Effect of morphine on the non-operated rats

3.3.1. Saline-pretreated group

In the saline-pretreated group (n = 4), 1 mg/kg of morphine produced a significant effect, lasting for 60 min with a maximum at 30 min ($P < 0.001$, paired t-test). At this time the mean threshold was 405 ± 16 g (pre-injection value = 285 ± 6 g).

3.3.2. Morphine-pretreated group

Similarly, in the morphine-pretreated group (n = 4) an acute injection of morphine produced a significant effect lasting for 60 min with a maximum at 30 min ($P < 0.001$, paired t-test). The maximal vocalization threshold was

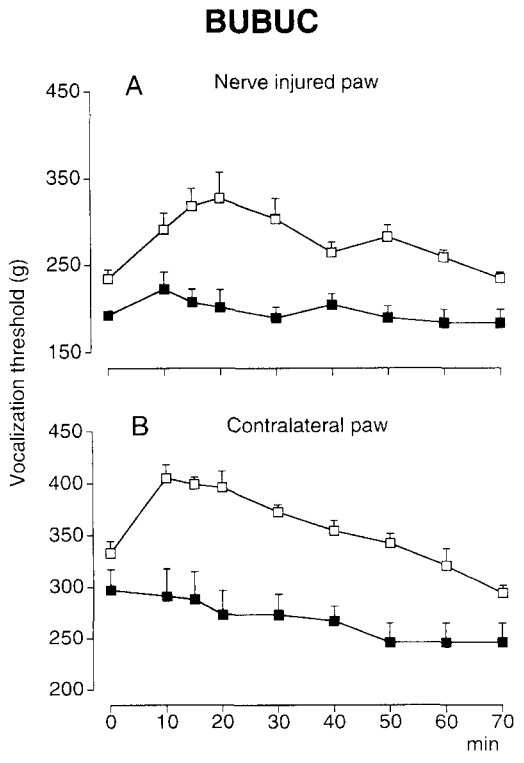

Fig. 3. The antinociceptive effect of BUBUC (3 mg/kg i.v.) on the vocalization threshold to paw pressure of the neuropathic rats pretreated with saline (□, n = 8) or morphine (■, n = 7). (A) nerve-injured paw, (B) contralateral paw.

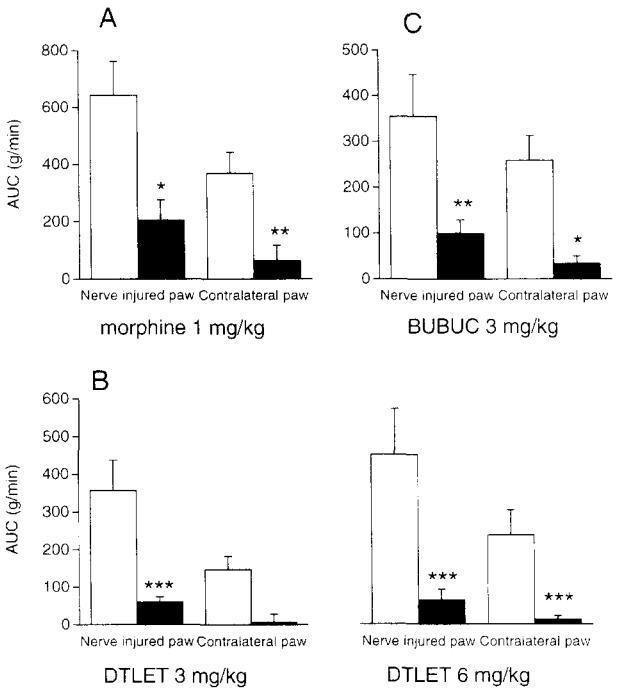

Fig. 4. Mean areas under the curves (AUC, g/min ± S.E.M.) of the antinociceptive time curves of morphine, BUBUC and DTLET in the paw pressure test of the nerve-injured and the contralateral paw. Open bars = pretreatment with saline; solid bars = pretreatment with morphine. *, $P < 0.05$, **, $P < 0.01$, ***, $P < 0.001$ (Fisher's PLSD test) compared with the saline-pretreated group.

414 ± 25 g (pre-injection value = 342 ± 14 g). There was no difference between the overall effects (AUCs) of morphine in the saline- and morphine-pretreated groups (N.S., unpaired t-test).

3.4. Effect of morphine in the neuropathic rats

3.4.1. Saline-pretreated group

In the saline-pretreated group, the effect of morphine on both hind paws was roughly comparable to that of the previous studies (Neil et al., 1990; Attal et al., 1991). Morphine (1 mg/kg) produced a significant effect on both hind paws, lasting for 70 min with a maximum at 30 min ($P < 0.001$, unpaired t-test, Fig. 1).

3.4.2. Morphine-pretreated group

In the morphine-pretreated group, the intensity and duration of the effect of morphine was strongly reduced (Fig. 1 A). The AUC for the nerve-injured paw fell to about a third of that of the saline-pretreated group (Fig. 4A). Morphine was ineffective on the contralateral paw (Fig. 1B and Fig. 4A).

3.5. Effect of DTLET and BUBUC in the neuropathic rats

3.5.1. Saline-pretreated group

In the saline-pretreated groups, DTLET and BUBUC produced moderate but significant effects on both hind

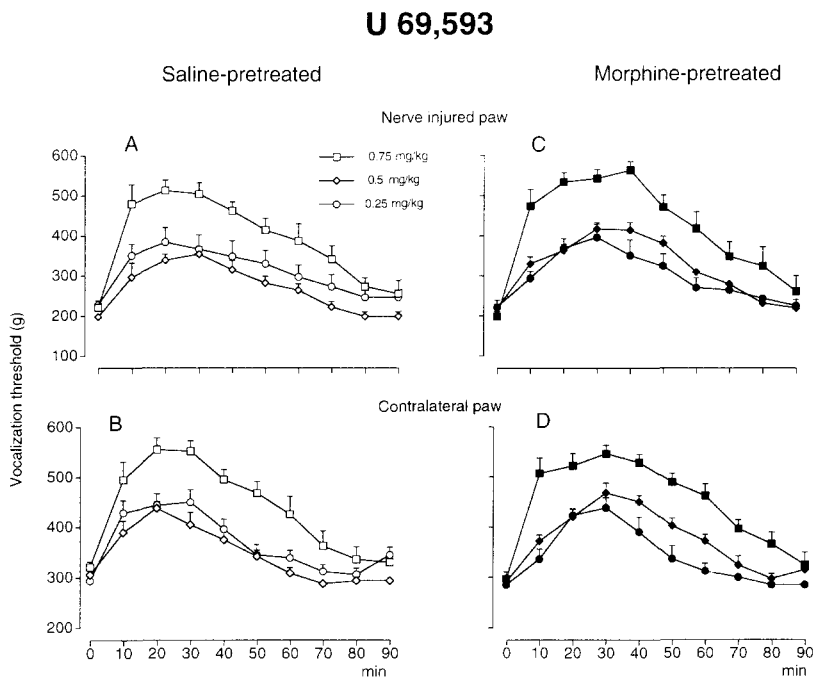

Fig. 5. The antinociceptive effect of U-69,593 (0.25, 0.5 and 0.75 mg/kg i.v.) on the vocalization threshold to paw pressure of the neuropathic rats pretreated with saline (□, ○, ◇, open symbols, 0.25 mg/kg $n = 6$, 0.5 mg/kg $n = 9$ and 0.75 mg/kg $n = 10$) or morphine (■, ●, ◆, 0.25 mg/kg $n = 6$, 0.5 mg/kg $n = 9$ and 0.75 mg/kg $n = 9$). A, C: nerve-injured paw; B, D: contralateral paw.

paws. The effect of 3 mg/kg DTLET on the nerve-injured paw, lasted for 60 min with a maximum at 20 min ($P < 0.001$, paired t-test; Fig. 2A). For the dose of 6 mg/kg of DTLET, the effect was significant on both hind paws (nerve-injured paw: $P < 0.001$, paired t-test; contralateral paw: $P < 0.05$, paired t-test; Fig. 2). With the dose of 3 mg/kg of BUBUC, the effect on both hind paws lasted for 70 min with a maximum at 20 min ($P < 0.001$, paired t-test; Fig. 3).

3.5.2. Morphine-pretreated group

In the morphine-pretreated group, the antinociceptive effects of both DTLET and BUBUC were abolished (Fig. 2 and Fig. 3). The AUCs for both hind paws were significantly smaller than those of the saline-pretreated group (Fig. 4B and C).

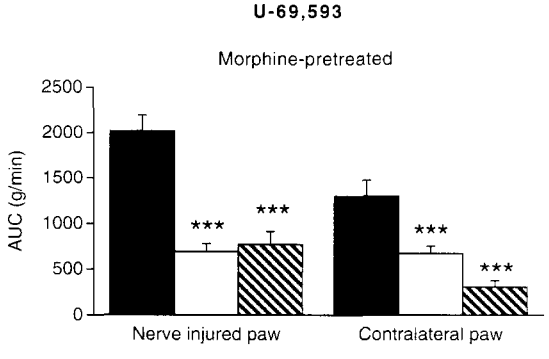

Fig. 6. Mean areas under the curves (AUC, g/min ± S.E.M.) of the antinociceptive time curves of the i.v. dose 0.75 mg/kg of U-69,593 alone (solid bars, $n = 9$) and the combination of U-69,593 (0.75 mg/kg i.v.) with the i.v. dose 1 mg/kg (open bars, $n = 6$) and 2 mg/kg (hatched bars, $n = 6$) of nor-binaltorphimine in the paw pressure test of the neuropathic rats pretreated with morphine. ***, $P < 0.001$ (Fisher's PLSD test) compared with the control group.

3.6. Effect of U-69,593 in the neuropathic rats

3.6.1. Saline-pretreated group

In the saline-pretreated group, U-69,593 induced a powerful, dose-dependent effect on both hind paws. With the dose 0.75 mg/kg of U-69,593, the effect lasted for 90 min and reached a maximum at 30 min ($P < 0.001$, paired t-test; Fig. 5A and B). Lower doses (0.25 and 0.5 mg/kg) of U-69,593 induced a less potent, but significant effect on both hindpaws, lasting for 80 min, with a maximum at 20 min ($P < 0.001$, paired t-test; Fig. 5A and B).

3.6.2. Morphine-pretreated group

In the morphine-pretreated group, the antinociceptive effect of U-69,593 was comparable to that found in the saline-pretreated group. With the dose 0.75 mg/kg of U-69,593, the antinociceptive effect lasted for 90 min with a maximum at 30 min ($P < 0.001$, paired t-test; Fig. 5C and D). The effect of U-69,593 (0.5 and 0.25 mg/kg) lasted for 80 min and reached a maximum at 20 min ($P < 0.001$, paired t-test; Fig. 5C and D). The AUCs for both hind paws were not significantly different from those of the saline-pretreated group (not shown).

The κ-opioid receptor antagonist, nor-binaltorphimine (1 and 2 mg/kg), significantly reversed the effect of 0.75 mg/kg of U-69,593 both on the nerve-injured (2/3 reduction) and on the contralateral paw (1/3 reduction for

nor-binaltorphimine 1 mg/kg; 2/3 reduction for nor-binaltorphimine 2 mg/kg; Fig. 6).

4. Discussion

In the present study, we clearly confirmed that systemically administered morphine and selective opioid receptor agonists are able to alleviate the abnormal responses to mechanical stimuli of the rats with peripheral mononeuropathy (Neil et al., 1990; Attal et al., 1991; Desmeules et al., 1993a). Irrespective of the opioid used, peak vocalization thresholds following drug administration were comparable between nerve-injured and contralateral paws, despite of lower pre-drug vocalization thresholds in the nerve-injured paw (Figs. 1–5). We have recently proposed that a contributory mechanism to the enhanced effect found in the nerve-injured paw, could be a peripheral action of the opioid receptor agonists on the nerve-injured paw (Kayser et al., 1995b; Catheline et al., 1995).

4.1. Tolerance to the antinociceptive effect of morphine in the neuropathic rats

In neuropathic animals, morphine pretreatment reduced the effect of acute morphine by about 3-fold for the nerve-injured, and abolished all effects on the contralateral paw. Although we did not assess the magnitude of tolerance by determining the rightward shift of the ED50, these findings clearly indicate that tolerance to the antinociceptive effect of morphine had developed. Since morphine was effective in non-operated rats handled in the same manner, there is no reason to suspect that environmental factors such as cues (Siegel, 1976; Wiertelak et al., 1992) contribute to the observed tolerance. It should also be noted that discrete signs of morphine withdrawal: piloerection, diarrhea and loss of body weight were observed in neuropathic rats. These signs have been used as an index of the intensity of physical dependence in morphine-treated rodents (Stolerman et al., 1975; Idänpään-Heikkilä et al., 1996). These results are not surprising, since in a preliminary study (Neil et al., 1990) in a small number of rats, we observed a decrease in the effect of morphine after the same pretreatment.

Our data may appear to be in contradiction to those of Backonja et al. (1995), who concluded that tolerance to morphine did not occur in neuropathic rats. However, the experimental procedure in this previous study was different: they used a different test (paw withdrawal to heat) and continuous infusion of morphine. The physiological state associated with continuous systemic infusion of morphine is quite distinct from that associated with repeated injections of the opioid. It has been reported that continuous infusion of morphine increased, while repeated injections of morphine decreased the levels of dynorphin in the rat pituitary gland (Rattan et al., 1992; Nylander et al., 1995). In addition, continuous infusion of morphine reduces, whereas repeated morphine injections enhance, dopaminergic neurotransmission in the rat striatum (Tjon et al., 1994). Furthermore, in the study of Backonja et al., 1995 a progressive loss of morphine analgesia was observed in neuropathic rats receiving 10–20 mg/kg morphine sulfate per day. This effect was claimed to be attributed to the progression of the disease but in our opinion, the development of some degree of tolerance to morphine cannot be excluded.

There is recent evidence that chronic administration of morphine can lead to a hyperalgesic state (Mao et al., 1994, Mao et al., 1995). It has also been shown previously that the baseline vocalization thresholds of the neuropathic rats are reduced by repeated administration of low doses of morphine without development of tolerance to morphine (Neil et al., 1990). Likewise, in our study, a decrease of the vocalization threshold was observed following the morphine pretreatment. It could be suggested that this decrease partially masked the effect of acute morphine in mononeuropathic rats. However, even if this pretreatment also lowered the vocalization threshold of the non-operated rats, it did not modify the effect of acute morphine in these animals. Therefore, it seems unlikely that the decrease in the threshold of the neuropathic rats could partially mask the effect of acute morphine and result in the loss of the efficacy of morphine in these animals.

Our recent experiments on sham operated rats (unpublished results) and the present results on unoperated rats show that in these animals, tolerance is not induced by the same morphine pretreatment that induces a clear tolerance to the neuropathic rats. Our findings are in agreement with previous studies performed in another model of persistent pain, the arthritic rat. These earlier studies showed that repeated morphine administration, high doses (starting at 40 mg/kg per day) (Kayser and Guilbaud, 1984) as well as low doses (0.3–3 mg/kg s.c. twice daily) (Kayser et al., 1986), induces tolerance to a greater extent in polyarthritic rats than in normal animals. Recent pieces of evidence suggest that in persistent pain in general, and in neuropathic pain in particular, the neuronal substrates of nociceptive information processing undergo significant plastic and biochemical changes (Stevens et al., 1991; Besse et al., 1992; Coderre et al., 1993). It is therefore not surprising that rats experiencing persistent pain respond to opioid treatment differently from normal animals in which the nociceptive circuitry has not undergone such modifications.

4.2. Loss of the efficacy of DTLET and BUBUC in morphine-tolerant neuropathic rats

In the present study, the antinociceptive effect produced by the δ-opioid receptor agonist DTLET was abolished both ipsi- and contra-laterally to the lesion in the morphine-pretreated neuropathic rats. It should be noted that a

similar loss of effect was observed with the other δ-opioid receptor agonist BUBUC which displays a better affinity and higher selectivity for δ-opioid receptors than DTLET (Delay-Goyet et al., 1991). We initially demonstrated that the antinociceptive effects of BUBU were prevented by the δ-opioid receptor antagonist naltrindole (Desmeules et al., 1993a,b), while they were not affected by naloxone, the μ-opioid receptor preferring antagonist, at a dose able to reverse the analgesic effect of the same order induced by morphine. These results suggest that the antinociceptive actions of BUBU are more likely to be mediated by δ- than μ-opioid receptors. Since a ceiling effect was observed with the doses 3 and 6 mg/kg of BUBUC in neuropathic rats (Desmeules et al., 1993a), higher doses of the agonist were not tested in the present study.

Compared to morphine, the magnitude of the effects obtained with DTLET and BUBUC was low. Indeed, when compared with μ-opioid receptor agonists, δ-opioid receptor agonists have generally been less effective (Stewart and Hammond, 1993). Furthermore, δ-opioid receptor agonists have been shown to be more effective in attenuating responses to noxious thermal than mechanical stimuli (Rodriguez et al., 1986).

The diminished effect of the δ-opioid receptor agonists in the neuropathic rats rendered tolerant to morphine, suggests that there is an interplay between μ- and δ-opioid receptors. In fact, there is previous evidence of an interaction between μ- and δ-opioid receptor types (Chang and Cuatrecasas, 1979; Vaught and Takemori, 1979; Lee et al., 1980; Rothman et al., 1985; Schoffelmeer et al., 1990; Porreca et al., 1992). Our findings are in agreement with previous studies showing that selective blockade of δ-opioid receptors prevents the development of morphine tolerance or dependence in rodents (Abdelhamid et al., 1991; Suzuki et al., 1995). Furthermore, δ-opioid receptor agonists have been shown to attenuate the development and expression of the signs of naloxone precipitated morphine abstinence syndrome (Lee et al., 1993).

4.3. Antinociceptive effect of U-69,593 in morphine-tolerant neuropathic rats

In our study, there was no change in the antinociceptive effect produced by the κ-opioid receptor agonist U-69,593 in the morphine-pretreated mononeuropathic rats as compared with the saline-pretreated animals. The effect of the κ-opioid receptor agonist in neuropathic rats was dose-dependent and reversed by the κ-selective opioid receptor antagonist nor-binaltorphimine. It should be noted that as previously shown (Lee et al., 1994), nor-binaltorphimine (1 mg/kg) alone has no significant effect in this model. These data indicate that the effect of U-69,593 in the morphine-pretreated neuropathic rats is unambiguously related to the activation of κ-opioid receptors.

Several studies have been undertaken to determine if there is a communication between the μ- and κ-opioid receptors. Both natural (Schmauss and Hertz, 1987; Tulunay et al., 1981) and synthetic κ-opioid receptor agonists (Ramarao et al., 1988; Picker et al., 1991) modulate morphine analgesia differently in morphine-naive and morphine-tolerant rodents. Furthermore, it has been shown that the intensity of dynorphin A-(1–13), an endogenous κ-opioid receptor agonist-induced catalepsy is greater in sufentanil (a μ-opioid receptor agonist)-tolerant rats than in non-tolerant rats (Herman and Goldstein, 1981). In the present study, side effects, such as catalepsy, which is induced by higher doses of U-69,593 in morphine-naive neuropathic rats (Desmeules et al., 1993a) were not noted. The present results thus indicate that the morphine pretreatment was unable to modify the κ-opioid receptor mediated effects. These data fit well with the receptor specificities of morphine and U-69,593. Morphine has the highest affinity to μ-opioid receptors, a much weaker affinity to the δ-opioid receptors, and only a weak affinity to κ-opioid receptors (Pasternak, 1993), whereas the κ-opioid receptor agonist U-69,593 is highly specific for receptors of the κ_1-opioid subtype. The persistence of the effect of U-69,593 in morphine-pretreated neuropathic rats is in agreement with a previous study where the density of the central κ_1-opioid receptors remained unaltered in morphine tolerant animals (Bhargava et al., 1991).

Our results are in further accordance with previous studies in which no cross-tolerance between μ- and κ-selective opioid receptor agonists has been observed (Craft and Dykstra, 1990; Bhargava et al., 1991; Kolesnikov et al., 1993; Elliott et al., 1994). In fact, although similarities between μ- and κ-opioid receptor agonists exist with respect to their ability to produce analgesia, there are many differences in their mechanisms of action (Cherubini and North, 1985; Reisine and Bell, 1993; Windh et al., 1995).

To conclude, it appears that the rats with peripheral mononeuropathy are exceptionally sensitive to morphine-induced tolerance. Morphine-tolerant neuropathic rats are also tolerant to the antinociceptive effects of δ-opioid receptor agonists and there is a lack of cross-tolerance between μ- and κ-selective opioid receptor agonists in neuropathic rats. Although the present results were obtained in a model of experimental neuropathic pain, they suggest again that κ-opioid receptor agonists may offer an additional therapeutic option for the alleviation of some clinical symptoms (such as mechano-allodynia) of peripheral neuropathic conditions. The possible use of such opioid receptor agonists in the treatment of neuropathic pain should be further explored.

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