Neuroscience Letters 562 (2014) 28–33

Contents lists available at ScienceDirect

Neuroscience Letters

journal homepage: www.elsevier.com/locate/neulet

The analgesic effect of tramadol in animal models of neuropathic pain and fibromyalgia

Kumi Kaneko*, Masato Umehara[1], Takashi Homan[1], Ken Okamoto[1], Michiko Oka, Tatsuya Oyama

Research Laboratories, Nippon Shinyaku Co., Ltd., 14, Nishinosho-monguchi-cho, Kisshoin, Minami-ku, Kyoto 601-8550, Japan

HIGHLIGHTS

- We compared the analgesic effects of tramadol in two kinds of chronic pain models.
- Oral treatment of tramadol improved partial sciatic nerve ligation-induced allodynia.
- Orally administered this drug also attenuated reserpine-induced tactile allodynia.
- The opioid system may be partly involved in the effect of tramadol on allodynia.

ARTICLE INFO

Article history:
Received 7 October 2013
Received in revised form
17 December 2013
Accepted 6 January 2014

Keywords:
Tramadol
Opioid
Neuropathic pain
Fibromyalgia
Partial sciatic nerve ligation
Reserpine
Naloxone

ABSTRACT

(±)-Tramadol hydrochloride (tramadol) is a widely used analgesic for the treatment of cancer pain and chronic pain. Although many animal studies have shown antinociceptive effects of tramadol in both acute and chronic pain, little is known about the effect of tramadol in putative animal models of fibromyalgia. In this study, we compared the antiallodynic effects of oral administration of tramadol in two kinds of rat chronic pain models, neuropathic pain induced by partial sciatic nerve ligation (PSL) and reserpine-induced myalgia (RIM). In PSL rats, the threshold for responses induced by tactile stimulation with von Frey filaments was significantly decreased seven days after the operation, suggesting that the operation induced tactile allodynia. Orally administered tramadol showed a potent and dose-dependent antiallodynic effect on PSL-induced allodynia. In RIM rats, the threshold was significantly decreased five days after reserpine treatment. Orally administered tramadol also attenuated reserpine-induced tactile allodynia. To explore the mechanism of the antiallodynic effect of tramadol in RIM rats, we investigated the effect of the opioid antagonist naloxone on the tramadol-induced analgesic effect in these rats. The effect of tramadol was partially antagonized by naloxone, suggesting that the opioid receptor is involved at least in part in the antiallodynic effect of tramadol in RIM rats. These data indicate that orally administered tramadol produced improvement in both PSL rats and RIM rats at similar doses and provide evidence that the opioid system is partly involved in the analgesic effect of tramadol in RIM rats.

1. Introduction

Neuropathic pain is a chronic condition characterized by spontaneous burning pain, hyperalgesia, and allodynia and it is very difficult to manage [1]. To clarify its mechanisms and develop effective therapies, several potential animal models of neuropathic pain have been developed and studied [2]. Partial sciatic nerve ligation (PSL) is a well characterized rat model of neuropathic pain with sciatic nerve injury, and it exhibits tactile allodynia [3].

Fibromyalgia is a musculoskeletal disorder characterized by chronic widespread pain and various comorbid symptoms such as depression. Although the details of its pathophysiology are unknown, biogenic amine levels in the cerebrospinal fluid are significantly lower than normal in fibromyalgia patients, suggesting dysfunction of the descending analgesic neural pathway [4]. Because there are few consistently effective therapies for fibromyalgia, more-effective agents are eagerly awaited. Several potential animal models have been described [5–9], and a reserpine-induced myalgia (RIM) model that may mimic various aspects of fibromyalgia has recently been reported [8]. In the RIM model, reserpine induces long-lasting muscle hyperalgesia and tactile allodynia and markedly decreases monoamine levels in the

Abbreviations: PSL, partial sciatic nerve ligation; RIM, reserpine-induced myalgia.

* Corresponding author. Tel.: +81 75 321 9179; fax: +81 75 314 3269.
E-mail address: k.kaneko@po.nippon-shinyaku.co.jp (K. Kaneko).

[1] These three authors contributed equally.

0304-3940/$ – see front matter © 2014 Elsevier Ireland Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.neulet.2014.01.007

spinal cord and some regions of the brain. In addition, there is an increase in the immobility time in the forced-swim test, an indicator of depression, which is a frequent comorbid symptom in fibromyalgia patients.

(±)-Tramadol hydrochloride (tramadol) is a widely used analgesic agent [10] that stimulates the μ-opioid receptor and inhibits serotonin and noradrenaline reuptake [11,12]. There are numerous animal studies on the antinociceptive effects of tramadol on heat pain [13] and chemical pain [14]. We have also recently demonstrated the effect of tramadol on visceral pain in rodent cystitis models [15]. However, few studies have been undertaken to investigate the effect of tramadol in putative experimental models of fibromyalgia. Therefore, we compared the mode of action of orally administered tramadol in PSL and RIM rats and investigated the mechanism of tramadol in RIM rats.

2. Materials and methods

2.1. Animals

Male Sprague–Dawley rats (aged 5–6 weeks for PSL and 9 weeks for RIM at the beginning of the experiment; Japan SLC, Hamamatsu, Japan) were housed under controlled environmental conditions (23 ± 3 °C; 12 h:12 h light–dark cycle, lights on at 08:00 h; free availability of food and water) for at least one week before use. The study was conducted in compliance with the Law for the Humane Treatment and Management of Animals (Law No. 105, 1 October 1973, as revised on 1 June 2006). All efforts were made to minimize animal suffering and to reduce the number of animals used.

2.2. Drugs

Tramadol was kindly supplied by Grünenthal (Aachen, Germany). Reserpine and naloxone hydrochloride were purchased from Wako Pure Chemical Industries (Osaka, Japan) and Tocris Bioscience (Bristol, UK), respectively. Reserpine was dissolved in glacial acetic acid, diluted with distilled water to a final concentration of 0.5% acetic acid, and injected subcutaneously in a volume of 1 ml/kg. Naloxone was dissolved in saline and administered subcutaneously in 1 ml/kg and tramadol was dissolved in distilled water and administered intraperitoneally in 1 ml/kg or orally in 5 ml/kg.

2.3. PSL surgery

PSL surgery was performed according to the method of Seltzer et al. [3]. Animals were anesthetized with pentobarbital. The right common sciatic nerve was exposed just distal to the branch leading to the posterior biceps femoris/semitendinosus muscle. The dorsal one-third to one-half of the sciatic nerve was tightly ligated with 8–0 silk suture and the wound was closed by suturing the muscle and skin layers. After recovering from the anesthesia, almost all animals showed guarding of the hind paw, but none engaged in autotomy. In sham-operated rats, the nerve was exposed but not ligated. To check the validity of the models, the tactile-response thresholds of all 10 PSL-operated rats were compared with those of five sham-operated rats seven days after surgery. The predrug tactile-response thresholds were measured first, and rats with a threshold of less than 5 g were selected for use in pharmacological tests as successful model rats. The selected rats were then allocated to four groups (n = 5 or 10/group), each of which received vehicle or tramadol. The tactile-response thresholds were again measured 1, 2 and 4 h after administration of vehicle or tramadol.

2.4. Reserpine-induced myalgia

RIM was produced in rats according to a previously reported protocol [8,16]. Briefly, reserpine was subcutaneously injected at a dose of 1 mg/kg once daily for three consecutive days. According to a previous study, food pellets were put on the floor of the cage after the reserpine injection as a nursing treatment so that the animals could access them more easily [16]. The baseline and time-course measurements of the tactile responses of seven RIM rats were compared with those of six sham-treated rats 1, 4, 7, 11 and 14 days after the last injection of reserpine. The effect of tramadol was evaluated five days after the last injection of reserpine. The predrug tactile-response thresholds were measured first and rats with a threshold of less than 5 g were selected for use in pharmacological tests as successful model rats. The selected rats were then allocated to four groups (n = 10/group), each of which received vehicle or tramadol. The tactile-response thresholds were again measured 1, 2 and 4 h after administration of vehicle or tramadol.

2.5. von Frey hair test

Tactile allodynia was measured by the up-down method as described previously [17]. Animals were individually placed on a wire-mesh floor and acclimatized to the environmental conditions for at least 30 min. After acclimatization, a tactile stimulus was applied to the middle plantar surface of the paw by placing one of a series of von Frey filaments (0.4, 0.6, 1.0, 2.0, 4.0, 6.0. 8.0 and 15.0 g) perpendicular to the surface of the paw. Testing was initiated with the 2.0-g filament. If this stimulus did not evoke a paw-withdrawal response, a stronger stimulus was presented; in the event of paw withdrawal, the next weaker stimulus was presented. Four additional responses were observed after the first response that changed from negative to positive or from positive to negative, and the 50% withdrawal threshold was determined. When continuous positive or negative responses were observed to the exhaustion of the stimulus set, values of 0.4 or 15.0 g, respectively, were assigned.

2.6. Statistical analysis

Data were analyzed by using SAS version 9.1.3 (SAS Institute, Cary, NC, USA) and were expressed as the mean ± standard error (SEM). Differences in the tactile-response threshold between PSL or RIM rats and sham-treated rats and between rats treated with saline/tramadol and rats treated with naloxone/tramadol were analyzed for statistical significance by the Wilcoxon rank-sum test. Differences in the threshold between rats before and after naloxone/tramadol treatment were analyzed for statistical significance by the Wilcoxon signed-rank test. To evaluate the effect of tramadol on the threshold, differences between the vehicle-treated and tramadol-treated groups were analyzed for statistical significance by the Shirley–Williams test. P-values less than 0.05 were considered significant.

3. Results

3.1. Effect of tramadol on tactile allodynia in PSL rats

We measured the withdrawal threshold of the ipsilateral paw seven days after PSL surgery and found that the mean threshold was markedly less than that of sham-operated rats (Fig. 1A), providing evidence that the operation had induced tactile allodynia. Intraperitoneally injected tramadol (10 and 30 mg/kg) significantly increased the mean threshold in a dose-dependent manner at 1 h after administration (Fig. 1B). Orally administered tramadol at the