Available online at www.sciencedirect.com

SCIENCE DIRECT

European Journal of Pharmacology 511 (2005) 21–26

www.elsevier.com/locate/ejphar

Role of 5-HT_{1A} and 5-HT_{1B} receptors in the antinociceptive effect of tramadol

M. Olga Rojas-Corrales, Esther Berrocoso, Juan A. Micó*

Pharmacology and Neuroscience Research Group (CTS-510), Department of Neuroscience, Faculty of Medicine, University of Cádiz, Plaza Fragela 9, 11003-Cádiz, Spain

Received 21 October 2004; received in revised form 27 January 2005; accepted 1 February 2005

Abstract

Tramadol, (1*RS*,2*RS*)-2-[(dimethylamine)-methyl]-1-(3-methoxyphenyl)-cyclohexanol hydrochloride, is an atypical centrally acting analgesic agent with relatively weak opioid receptor affinity and which, like some antidepressants, is able to inhibit the reuptake of serotonin (5-hydroxytryptamine, 5-HT) in the raphe nucleus. We have previously demonstrated that pindolol, a beta-adrenoceptor blocker/5-hydroxytryptamine$_{1A/1B}$ receptor antagonist, enhanced tramadol antinociception and that the selective 5-HT_{1A} agonist 8-Hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT) reduced it. These effects were related to the negative feedback control that regulates raphe region neurones. The current study examines the ability of the selective antagonist at somatodendritic 5-HT_{1A} receptors, *N*-{2-[4-(2-methoxyphenyl)-1-piperazinyl] ethyl}-*N*-(2-pyridinyl) cyclohexane carboxamide (WAY100635, 0.8 mg/kg), the selective antagonist at terminal 5-HT_{1B} receptors, *N*-[3-(2-dimethylamino) ethoxy-4-methoxyphenyl]-2′-methyl-4′-(5-methyl-1,2,4-oxadiazol-3-yl)-(1,1′-biphenyl)-4-carboxamide (SB216641, 0.1–0.8 mg/kg) and the selective agonist at 5-HT_{1B} receptors, 1,4-tDihydro-3-(1,2,3,6-tetrahydro-4-pyridinyl)-5*H*-pyrrolo[3,2-*b*] pyridin-5-one (CP93129, 0.2–0.4 mg/kg), to modify the antinociceptive effect of 4–64 mg/kg of tramadol in the hot plate test in mice. The results show that 0.8 mg/kg of WAY100635 enhanced antinociceptive effect of tramadol while neither agonism nor antagonism at the 5-HT_{1B} receptor modifies it significantly at the doses tested. These results account for involvement of the somatodendritic 5-HT_{1A} receptors in the analgesic effect of tramadol and support the supraspinal interaction of serotonin and the opioid system in the regulation of pain.

Keywords: Tramadol; Serotonin; Selective 5-HT_{1A}/5-HT_{1B} receptor antagonist; Antinociception; Hot plate; (Mice)

1. Introduction

Tramadol, (1*RS*,2*RS*)-2-[(dimethylamine)-methyl]-1-(3-methoxyphenyl)-cyclohexanol hydrochloride, is a centrally acting analgesic which has long been used in clinical practice. It is a synthetic opioid in the aminocyclohexanol group (Codd et al., 1995) that binds weakly but effectively to μ-opioid receptors (Hennies et al., 1988). Nevertheless, a non-opioid mechanism is also involved in tramadol analgesia, which consists in an enhancement of the extraneuronal concentration of noradrenalin and serotonin (5-hydroxytryptamine, 5-HT) by interference with both the uptake and release mechanisms (Bamigbade et al., 1997; Driessen and Reimann, 1992; Driessen et al., 1993; Raffa et al., 1992). This dual mechanism could be attributed to the two isomers found in the racemic mixture, which increase the concentration of these two neurotransmitters with different affinities in selected brain areas (Fink et al., 1970).

These monoaminergic properties, together with tramadol's chemical structure, both similar to that of some antidepressants (Markowitz and Patrick, 1998), may also account for other effects induced by tramadol. Interaction of the opioid and monoaminergic systems has been proposed as beneficial in the relief of severe depression (Schreiber et al., 2002). Therefore, it is not surprising that tramadol has shown antidepressant-like effect in behavioral models in

* Corresponding author. Tel.: +34 956 015 247; fax: +34 956 015 225.
E-mail address: juanantonio.mico@uca.es (J.A. Micó).

0014-2999/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.ejphar.2005.02.006

mice (Rojas-Corrales et al., 1998) and rats (Rojas-Corrales et al., 2002). Moreover, opioids with serotoninergic properties, including tramadol, have been proven to induce antidepressant-like effects in a classical model of depression (Rojas-Corrales et al., 2004). Furthermore, in the clinical context, tramadol has been successfully used in several psychiatric situations: in refractory major depression (Shapira et al., 2001), in severe suicidal ideation (Spencer, 2000), in antidepressant potentiation (Fanelli and Montgomery, 1998) and in the initial treatment of obsessive-compulsive disorder (Goldsmith et al., 1999; Shapira et al., 1997).

It is widely accepted that an enhancement of 5-HT levels is subject to receptor mediated autoregulatory control at the somatodendritic level in the raphe region. Acutely administered selective serotonin reuptake inhibitors preferentially increase extracellular levels of serotonin in the raphe nuclei, resulting in an activation of somatodendritic 5-HT_{1A} receptors and slowing the spontaneous firing rate of its neurones (Sprouse and Aghajanian, 1987). Thus, the preventive blocking of somatodendritic 5-HT_{1A} autoreceptors by specific receptor antagonists prevents the reduction of 5-HT release in terminal areas caused by 5-HT uptake blockers and potentiates their effects in terminal areas (Artigas, 1995; Romero and Artigas, 1997). In this respect, the time lag for the therapeutic effect of antidepressant drugs has been related to the down-regulation of 5-HT_{1A} autoreceptors (Chaput et al., 1986). Nevertheless, there is a growing body of evidence suggesting that further subtypes of 5-HT_1 receptors are involved in the regulation of the central serotonin neurotransmission, particularly 5-HT_{1B} and/or 5-HT_{1D} receptors (Hopwood and Stamford, 2001).

Tramadol, as well as selective serotonin reuptake inhibitors, enhances extracellular 5-HT levels in the vicinity of cell bodies and dendrites in the raphe nuclei (Bamigbade et al., 1997) and subsequently it may indirectly activate presynaptic 5-HT_1 receptors, bringing into action a negative feedback control. As raphe nuclei have been involved in pain modulation (Wang and Nakai, 1994), and a serotoninergic component has been demonstrated to be involved in tramadol analgesia (Oliva et al., 2002; Raffa et al., 1992), we are interested to study if this negative feedback control may curb the antinociceptive effect of tramadol. Previously, we have enhanced the antinociceptive effect of tramadol by co-administration of pindolol, a putative antagonist at β-adrenergic and 5-$HT_{1A/B}$ receptors and reduced by the selective agonist of 5-HT_{1A} receptors, 8-Hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT) (Rojas-Corrales et al., 2000). However, pindolol also blocks β-adrenoceptors and 5-HT_{1B} receptors, and a series of studies suggested that the clinical efficacy of pindolol may be unrelated to a restoration of serotoninergic neuronal activity in the dorsal raphe nucleus (Fornal et al., 1999). Furthermore, some authors have reported agonistic properties of pindolol at 5-HT_{1A} receptors (Clifford et al., 1998). Therefore, the aim of this study was to elucidate the role of 5-HT_{1A} and 5-HT_{1B} receptors in tramadol analgesia. Thus we set out to study both if selective blockade of 5-HT_{1A} or 5-HT_{1B} receptors, by means of selective receptor antagonists, modifies the antinociceptive effect of tramadol and if co-administration of a selective agonist of 5-HT_{1B} receptor also reduces the antinociceptive effect of tramadol to the same degree that 8-OH-DPAT does.

2. Materials and methods

2.1. Animals

Albino male mice of the CD1 strain (25–30 g) provided by the "Servicio de Experimentación y Producción Animal" (SEPA) of the University of Cádiz were used. Animals were maintained under standard conditions: 12-h light–dark schedule (light on at 8 h 00 min a.m.) with ad libitum food and water and constant temperature (21±1 °C). The experimental protocols were reviewed and approved by the Local Committee for Animal Experimentation of the Faculty of Medicine at the University of Cádiz (License number 079604) and complied with the International Association for the Study of Pain ethical guidelines (Zimmermann, 1983). Animal care and use procedures conformed to International European Ethical Standards (86/609-EEC) and Spanish Law (RD 223/1988).

2.2. Drugs

Tramadol, (1*RS*,2*RS*)-2-[(dimethylamine)-methyl]-1-(3-methoxyphenyl)-cyclohexanol hydrochloride, (Grünenthal-Andrómaco, Spain) was i.p. administered 30 min before the test. The selective somatodendritic 5-HT_{1A} receptor antagonist *N*-{2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl}-*N*-(2-pyridynyl) cyclohexane carboxamide (WAY100635, Sigma-Aldrich-Química, Spain), the selective 5-HT_{1B} receptor antagonist *N*-[3-(3-dimethylamino) ethoxy-4-methoxyphenyl]-2′-methyl-4′-(5-methyl-1,2,4-oxadiazol-3-yl)-(1,1′-biphenyl)-4-carboxamide (SB216641, Tocris, UK) and the selective 5-HT_{1B} agonist 1,4-tDihydro-3-(1,2,3,6-tetrahydro-4-pyridinyl)-5*H*-pyrrolo[3,2-*b*] pyridin-5-one (CP93129, Tocris, UK) were s.c. administered 15 min before the test. All the drugs were dissolved in saline (NaCl 0.9%). Control animals received saline only. Drugs were injected in a volume of 0.1 ml/100 g of body weight to the experimental groups (n=10).

2.3. Experimental procedure

Animals were placed on a hot plate apparatus (Digital DS-37 Socrel model) thermostatically maintained at 55.5±0.2 °C. Latency was the time elapsed either to lick or to flinch the hind paw or jumping. The latency to the first response of the animal (either paw licking or paw flinching or jumping) was recorded as the pain response latency in

seconds. Basal latency was recorded, being the mean value of two or three determinations before treatments. A cutoff time of 60 s was established. The dose–effect curves of tramadol alone and plus either WAY100635 or SB216641 were carried out simultaneously. Subsequently, dose–effect curves of tramadol and tramadol plus CP93129 were carried out in order to confirm the putative involvement of 5-HT_{1B} receptors in tramadol antinociception.

2.4. Statistical analysis

The results are expressed as percentages of maximal possible effect (%MPE) induced by the drugs related to basal latencies (%MPE=((test latency−basal latency)/(cutoff−basal latency))×100)±S.E.M. Data were analyzed using two-way analysis of variance, the two factors evaluated were: the doses of tramadol used to obtain the dose–response curves, and the drugs used (tramadol or tramadol plus WAY100635 or SB216641 or CP93129). The effect of each dose of tramadol was compared to that obtained with the combination (Student's *t*-test). A *P* value of <0.05 was considered to be significant.

3. Results

3.1. Effect of selective 5-HT_{1A} blockade in the antinociceptive effect of tramadol

Analysis of variance shows, as expected, a significant effect of 4–64 mg/kg of tramadol ($F_{(99,4)}$=72.950, P<0.001) in mouse nociception in the hot plate test, measured as percentage of maximal possible effect (%MPE). Administration of 0.8 mg/kg of WAY100635 also induced a significant effect ($F_{(99,1)}$=21.023, P<0.001) in tramadol-treated animals. No interaction of the two factors was observed ($F_{(99,4)}$=1.212, P=0.311). A *t*-test for no-paired samples shows that 0.8 mg/kg of WAY10035 enhanced significantly the effect induced by tramadol administered at

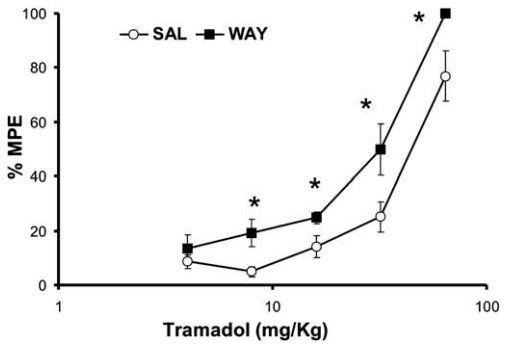

Fig. 1. Effect of the selective 5-HT_{1A} antagonist WAY 100635 (WAY) (0.8 mg/kg s.c., 15 min before test) in the antinociceptive effect of 4–64 mg/kg of tramadol (30 min before test) measured as percentage of maximum possible effect (%MPE)±S.E.M. in the hot plate test in mice. *P<0.05 vs. SAL (saline).

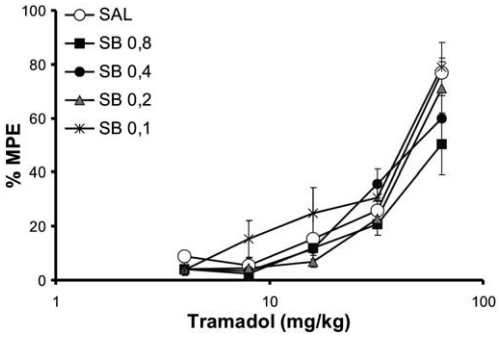

Fig. 2. Effect of the selective 5-HT_{1B} antagonist SB216641 (SB) (0.1–0.8 mg/kg s.c., 15 min before test) in the antinociceptive effect of 4–64 mg/kg of tramadol (i.p. 30 min before test) measured as percentage of maximum possible effect (%MPE)±S.E.M. in the hot plate test in mice. SAL: saline.

8 (19.18±4.92 vs. 4.88±1.85, P<0.05), 16 (24.85±2.24 vs. 14.15±4.01, P<0.05), 32 (49.80±9.49 vs. 25.09±5.54, P<0.05) and 64 (100.00 vs. 76.92±9.28, P<0.05) mg/kg. The effect of 4 mg/kg of tramadol was not modified by 0.8 mg/kg of WAY100635 (13.60±4.98 vs. 8.67±2.60, n.s.). No effect was observed with WAY100635 administered alone (data not shown) (Fig. 1).

3.2. Effect of selective 5-$HT_{1B/D}$ blockade in the antinociceptive effect of tramadol

Analysis of variance shows a significant effect of 4–64 mg/kg of tramadol ($F_{(99,4)}$=39.184, P<0.001) measured as percentage of maximal possible effect in the hot plate test. Co-administration of 0.8 mg/kg of SB216641 also induced a significant effect ($F_{(99,1)}$=5.321, P=0.023) in tramadol-treated mice. No interaction of the two treatments was observed ($F_{(99,4)}$=1.734, P=0.149). However, comparisons of groups shows that only the effect induced by 64 mg/kg of tramadol was slightly reduced (near statistical significance) by 0.8 mg/kg of SB216641 (50.50±11.25 vs. 76.92±9.29, P=0.087). Co-administration of 0.8 mg/kg of SB216641 to 4–32 mg/kg of tramadol induced percentages of maximal possible effect (3.95±1.87; 2.41±1.89; 11.87±3.93; 20.58±3.80, respectively) not significantly different from those induced by tramadol alone. Lower doses of SB216641 (0.1–0.4 mg/kg) did not show any effect on tramadol-induced antinociception. Administration of SB216641 0.1–0.8 mg/kg alone did not induce any effect on nociception (data not shown) (Fig. 2).

3.3. Effect of selective 5-$HT_{1B/D}$ agonism in the antinociceptive effect of tramadol

Administration of 0.2–0.4 mg/kg of CP93129 did not induce any significant effect ($F_{(149,2)}$=2.283, P=0.106) while 4–64 mg/kg of tramadol induced a significant effect on pain threshold measured as percentage of maximal possible effect ($F_{(149,4)}$=115.444, P<0.001). Co-administration of 0.2 and 0.4 mg/kg of CP93129 to 4–64 mg/kg of

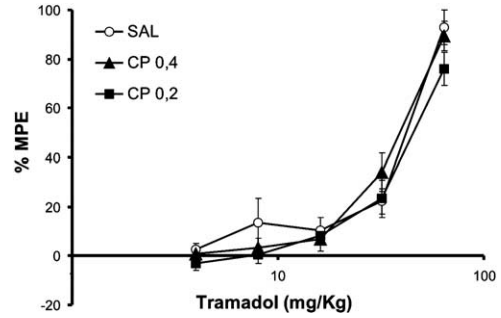

Fig. 3. Effect of the selective 5-HT_{1B} receptor agonist CP93129 (CP) (0.2–0.4 mg/kg, s.c., 15 min before test) in the antinociceptive effect of 4–64 mg/kg of tramadol (i.p. 30 min before test) measured as percentage of maximum possible effect (%MPE)±S.E.M. in the hot plate test in mice. SAL: saline.

tramadol did not induce any significant effect (−3.20±2.78 and 0.68±2.98 vs. 2.56±2.40, n.s.; 0.56±3.60 and 3.36±3.97 vs. 13.46±9.88, n.s.; 8.26±3.29 and 6.70±4.86 vs. 10.26±5.49, n.s.; 23.32±7.58 and 34.05±7.81 vs. 22.18±5.27, n.s.; 76.10±6.74 and 89.48±5.89 vs. 93.00±7.00, n.s.) Administration of CP93129 0.2–0.4 mg/kg alone did not induce any significant effect on pain threshold (data not shown) (Fig. 3).

4. Discussion

The role of opioid and monoamine systems in tramadol-induced analgesia is still not well known, but 5-HT has been demonstrated to play a role. In this study we investigated the involvement of two types of 5-HT_1 receptors in tramadol antinociception by means of selective receptor antagonists, and demonstrated that the selective antagonism of 5-HT_{1A} receptors enhances tramadol antinociception, while selective blockade or selective agonism of 5-HT_{1B} receptors did not induce a net effect. Opiate-like behavioral effects (as circling behavior and Straub sign) were observed at high doses of tramadol; however, the other drugs affecting 5-HT system did not affect these behaviors. Pharmacokinetic interactions were excluded because we have used these pharmacological combinations in other tests, no analgesic tests, and the effects were quite different.

Tramadol is an opioid analgesic with a pharmacodynamic profile distinct from the classical opioid analgesics. Tramadol induces antidepressant-like effects in rodents (Rojas-Corrales et al., 1998, 2002, 2004) and induces changes in CNS similar to that induced with conventional antidepressants; it decreases the binding of frontocortical β-adrenoceptors, 5-HT_{2A} receptors (Hopwood et al., 2001) and α_2-adrenoceptors (Faron-Gorecka et al., 2004a), whereas it increases the binding of α_1-adrenoceptors and dopamine D_2/D_3 receptors (Faron-Gorecka et al., 2004b). Moreover, tramadol-induced antinociception has been enhanced by pindolol (Rojas-Corrales et al., 2000), a strategy used to shorten the onset of action of systemic antidepressants (selective serotonin reuptake inhibitors) decreasing the inhibitory feedback of 5-HT neurons firing rate. This effect seemed to be related to blockade of 5-HT_{1A}, since the selective 5-HT_{1A} agonist 8-OH-DPAT reduced tramadol-induced antinociception.

A similar mechanism of negative feedback has been proposed regarding the noradrenergic system. Thus, blockade of α_2-receptors in the locus coeruleus induces an increase in noradrenalin levels at postsynaptic sites (Mateo et al., 1998). However, administration of α_2-receptor antagonists reduces tramadol antinociception, in both rodents and humans (Kayser et al., 1992, Desmeules et al., 1996). On the other hand, the antinociceptive effect of 8-OH-DPAT in the hot plate test has been antagonized by idazoxan, a α_2-receptor antagonist (Millan and Colpaert, 1991). Given these findings, further studies regarding supraspinal noradrenergic mechanisms involved in tramadol analgesia using selective ligands are needed.

Further, the mechanism of pindolol it is not well known. Some authors suggest that it may act as an agonist at 5-HT_{1A} receptors in the raphe nucleus (Clifford et al., 1998). Others have reported that pindolol may also act at terminal level, at the presynaptic 5-HT_{1B} serotoninergic receptors (Bourin et al., 1998). In fact, multiple 5-HT_1 subtypes govern different aspects of 5-HT function in the dorsal raphe nucleus, including 5-$HT_{1B/D}$ receptors (Stamford et al., 2000). Moreover the functional activity of the different subtypes of 5-HT_1 receptors may compensated one by another (Gardier et al., 2001). Very recently, it has been reported that 5-HT_{1A} and 5-HT_{1B} receptors play different roles in modulating the action of antidepressant drugs. Therefore blockade of 5-HT_{1B} rather than 5-HT_{1A} receptors may facilitate the effect of several antidepressants in the forced swimming test (Tatarczynska et al., 2004). Studies in knockout mice have shown that 5-HT_{1B} autoreceptors play a significant role in the inhibition of 5-HT release at serotoninergic nerve terminals in response to serotonin reuptake inhibitors (De Groote et al., 2002). However our study failed to demonstrate an involvement of 5-HT_{1B} receptor in tramadol-induced antinociception with 0.1–0.8 mg/kg of SB216641. The range of dose of SB216641 was chosen taking into account that the effective receptor antagonist concentration of this drug is very similar to that of WAY100635 (Stamford et al., 2000). However, the effect of administration of higher doses of SB21664 could also be studied.

Interaction of the serotoninergic and opioid systems has been documented in animal models concerning pain modulation (Schreiber et al., 1996). The exact mechanism by which 5-HT is involved in the effects of opioids has been questioned (Gao et al., 1998). It is well known that 5-HT plays a multifaceted role in the regulation of nociceptive transmission. Complexity arises from its actions at multiple sites within the pain transmission system (periphery, spinal cord, and supraspinal sites), and from actions by multiple 5-HT receptor subtypes. Our results are consistent with the

involvement of the 5-HT system in opioid-analgesia, probably via 5-HT_{1A} receptors preventing the reduction of 5-HT release in terminal areas. This process accounts for ascending 5-HT projections from the dorsal raphe nucleus and probably for descending serotoninergic projections from the magnus raphe nucleus to the dorsal horn. 5-HT_{1A} receptors are also located at the postsynaptic level, and play a complex role in analgesia. Bardin and Colpaert (2004) have demonstrated that opioids and 5-HT_{1A} receptor agonists can act and interact at spinal level to produce both hyper- and hypo-analgesic effects. Nevertheless, our results are concerned with a supraspinal involvement in pain modulation, in which mesencephalic nuclei in raphe region are highly involved (Wang and Nakai, 1994).

Our results are consistent with the hypotheses that serotonin system is closely involved in tramadol antinociception. A recent study has systematically reviewed the evidence from randomized controlled trials for the efficacy of tramadol in treating neuropathic pain (Duhmke et al., 2004), an effect usually related to antidepressant drugs. The involvement of 5-HT_1 receptors in this anti-neuropathic effect remains unclear. Deseure et al. (2002) reported a potent analgesic effect with systemic 5-HT_{1A} agonist in a model of trigeminal neuropathic pain. However, inhibition of 5-HT_{1A} expression (Hernandez et al., 2004), or blockade of 5-HT_{1A} receptors (Marchand et al., 2004), has been proposed as useful for treating neuropathic pain when combined with antidepressants. More in line with the findings reported in recent papers, our data support a counteracting role of somatodendritic 5-HT_{1A} receptors in the antinociceptive efficacy of tramadol and antidepressants with a serotoninergic spectrum, which may be of interest for new strategies for treating several pain conditions. Nevertheless, taking into account that the influence of 5-HT_{1A} ligands on nociception is dependent upon the algesiometric paradigm (Millan et al., 1996), further studies related to the involvement of different subtypes of 5-HT_1 receptors in the antinociceptive efficacy of tramadol are needed, especially in neuropathic pain models.

Acknowledgements

This study was supported by FIS-PI031430 and PAI-CTS510.

References

Artigas, F., 1995. Pindolol, 5-hydroxytryptamine, and antidepressant augmentation. Arch. Gen. Psychiatry 52, 969–971.

Bamigbade, T.A., Davidson, C., Langford, R.M., Stamford, J.A., 1997. Actions of tramadol, its enantiomers and principal metabolite, *O*-desmethyltramadol, on serotonin (5-HT) efflux and uptake in the rat dorsal raphe nucleus. Br. J. Anaesth. 79, 352–356.

Bardin, L., Colpaert, F.C., 2004. Role of spinal 5-HT(1A) receptors in morphine analgesia and tolerance in rats. Eur. J. Pain 8, 253–261.

Bourin, M., Redrobe, J.P., Baker, G.B., 1998. Pindolol does not act only on 5-HT1A receptors in augmenting antidepressant activity in the mouse forced swimming test. Psychopharmacology (Berl) 136, 226–234.

Chaput, Y., de Montigny, C., Blier, P., 1986. Effects of a selective 5-HT reuptake blocker, citalopram, on the sensitivity of 5-HT autoreceptors: electrophysiological studies in the rat brain. Naunyn-Schmiedeberg's Arch. Pharmacol. 333, 342–348.

Clifford, E.M., Gartside, S.E., Umbers, V., Cowen, P.J., Hajos, M., Sharp, T., 1998. Electrophysiological and neurochemical evidence that pindolol has agonist properties at the 5-HT1A autoreceptor in vivo. Br. J. Pharmacol. 124, 206–212.

Codd, E.E., Shank, R.P., Schupsky, J.J., Raffa, R.B., 1995. Serotonin and norepinephrine uptake inhibiting activity of centrally acting analgesics: structural determinants and role in antinociception. J. Pharmacol. Exp. Ther. 274, 1263–1270.

Deseure, K., Koek, W., Colpaert, F.C., Adriaensen, H., 2002. The 5-HT(1A) receptor agonist F 13640 attenuates mechanical allodynia in a rat model of trigeminal neuropathic pain. Eur. J. Pharmacol. 456, 51–57.

De Groote, L., Olivier, B., Westenberg, H.G., 2002. Extracellular serotonin in the prefrontal cortex is limited through terminal 5-HT(1B) autoreceptors: a microdialysis study in knockout mice. Psychopharmacology (Berl) 162, 419–424.

Desmeules, J.A., Piguet, V., Collart, L., Dayer, P., 1996. Contribution of monoaminergic modulation to the analgesic effect of tramadol. Br. J. Clin. Pharmacol. 41, 7–12.

Driessen, B., Reimann, W., 1992. Interaction of the central analgesic, tramadol, with the uptake and release of 5-hydroxytryptamine in the rat brain in vitro. Br. J. Pharmacol. 105, 147–151.

Driessen, B., Reimann, W., Giertz, H., 1993. Effects of the central analgesic tramadol on the uptake and release of noradrenaline and dopamine in vitro. Br. J. Pharmacol. 108, 806–811.

Duhmke, R.M., Cornblath, D.D., Hollingshead, J.R., 2004. Tramadol for neuropathic pain. Cochrane Database Syst. Rev. 2 (CD003726).

Fanelli, J., Montgomery, C., 1998. Use of the analgesic tramadol in antidepressant potentiation. Psychopharmacol. Bull. 32, 442.

Faron-Gorecka, A., Kusmider, M., Inan, S.Y., Siwanowicz, J., Dziedzicka-Wasylewska, M., 2004a. Effects of tramadol on alpha2-adrenergic receptors in the rat brain. Brain Res. 1016, 263–267.

Faron-Gorecka, A., Kusmider, M., Inan, S.Y., Siwanowicz, J., Piwowarczyk, T., Dziedzicka-Wasylewska, M., 2004b. Long-term exposure of rats to tramadol alters brain dopamine and alpha(1)-adrenoceptor function that may be related to antidepressant potency. Eur. J. Pharmacol. 501, 103–110.

Fink, M., Simeon, J., Itil, T.M., Freedman, A.M., 1970. Clinical antidepressant activity of cyclazocine—a narcotic antagonist. Clin. Pharmacol. Ther. 11, 41–48.

Fornal, C.A., Martin, F.J., Mendlin, A., Metzler, C.W., Bjorvatn, B., Jacobs, B.L., 1999. Pindolol increases extracellular 5-HT while inhibiting serotonergic neuronal activity. Eur. J. Pharmacol. 377, 187–191.

Gao, K., Chen, D.O., Genzen, J.R., Mason, P., 1998. Activation of serotonergic neurons in the raphe magnus is not necessary for morphine analgesia. J. Neurosci. 18, 1860–1868.

Gardier, A.M., Gruwez, B., Trillat, A.C., Jacquot, C., Hen, R., Bourin, M., 2001. Interaction between 5-HT(1A) and 5-HT(1B) receptors: effects of 8-OH-DPAT-induced hypothermia in 5-HT(1B) receptor knockout mice. Eur. J. Pharmacol. 421, 171–175.

Goldsmith, T.B., Shapira, N.A., Keck Jr., P.E., 1999. Rapid remission of OCD with tramadol hydrochloride. Am. J. Psychiatry 156, 660–661.

Hennies, H.H., Friderichs, E., Schneider, J., 1988. Receptor binding, analgesic and antitussive potency of tramadol and other selected opioids. Arzneim.-Forsch. 38, 877–880.

Hernandez, A., Constandil, L., Laurido, C., Pelissier, T., Marchand, F., Ardid, D., Alloui, A., Eschalier, A., Soto-Moyano, R., 2004. Venlafaxine-induced depression of wind-up activity in mononeuropathic rats is potentiated by inhibition of brain 5-HT1A receptor expression in vivo. Int. J. Neurosci. 114, 229–242.

Hopwood, S.E., Stamford, J.A., 2001. Multiple 5-HT(1) autoreceptor subtypes govern serotonin release in dorsal and median raphe nuclei. Neuropharmacology 40, 508–519.

Hopwood, S.E., Owesson, C.A., Callado, L.F., McLaughlin, D.P., Stamford, J.A., 2001. Effects of chronic tramadol on pre- and post-synaptic measures of monoamine function. J. Psychopharmacol. 15, 147–153.

Kayser, V., Besson, J.M., Guilbaud, G., 1992. Evidence for a noradrenergic component in the antinociceptive effect of the analgesic agent tramadol in an animal model of clinical pain, the arthritic rat. Eur. J. Pharmacol. 224, 83–88.

Marchand, F., Pelissier, T., Eschalier, A., Ardid, D., Alloui, A., Soto-Moyano, R., Mondaca, M., Laurido, C., Constandil, L., Hernandez, A., 2004. Blockade of supraspinal 5-HT1A receptors potentiates the inhibitory effect of venlafaxine on wind-up activity in mononeuropathic rats. Brain Res. 1008, 288–292.

Markowitz, J.S., Patrick, K.S., 1998. Venlafaxine–tramadol similarities. Med. Hypotheses 51, 167–168.

Mateo, Y., Pineda, J., Meana, J.J., 1998. Somatodendritic alpha2-adrenoceptors in the locus coeruleus are involved in the in vivo modulation of cortical noradrenaline release by the antidepressant desipramine. J. Neurochem. 71, 790–798.

Millan, M.J., Colpaert, F.C., 1991. Alpha 2 receptors mediate the antinociceptive action of 8-OH-DPAT in the hot-plate test in mice. Brain Res. 539, 342–346.

Millan, M.J., Seguin, L., Honore, P., Girardon, S., Bervoets, K., 1996. Pro- and antinociceptive actions of serotonin (5-HT)1A agonists and antagonists in rodents: relationship to algesiometric paradigm. Behav. Brain Res. 73, 69–77.

Oliva, P., Aurilio, C., Massimo, F., Grella, A., Maione, S., Grella, E., Scafuro, M., Rossi, F., Berrino, L., 2002. The antinociceptive effect of tramadol in the formalin test is mediated by the serotonergic component. Eur. J. Pharmacol. 445, 179–185.

Raffa, R.B., Friderichs, E., Reimann, W., Shank, R.P., Codd, E.E., Vaught, J.L., 1992. Opioid and nonopioid components independently contribute to the mechanism of action of tramadol, an 'atypical' opioid analgesic. J. Pharmacol. Exp. Ther. 260, 275–285.

Rojas-Corrales, M.O., Gibert-Rahola, J., Mico, J.A., 1998. Tramadol induces antidepressant-type effects in mice. Life Sci. 63, PL175–PL180.

Rojas-Corrales, M.O., Ortega-Alvaro, A., Gibert-Rahola, J., Roca-Vinardell, A., Mico, J.A., 2000. Pindolol, a beta-adrenoceptor blocker/5-hydroxytryptamine(1A/1B) antagonist, enhances the analgesic effect of tramadol. Pain 88, 119–124.

Rojas-Corrales, M.O., Berrocoso, E., Gibert-Rahola, J., Mico, J.A., 2002. Antidepressant-like effects of tramadol and other central analgesics with activity on monoamines reuptake, in helpless rats. Life Sci. 72, 143–152.

Rojas-Corrales, M.O., Berrocoso, E., Gibert-Rahola, J., Mico, J.A., 2004. Antidepressant-like effect of tramadol and its enantiomers in reserpinized mice: comparative study with desipramine, fluvoxamine, venlafaxine and opiates. J. Psychopharmacol. 18, 404–411.

Romero, L., Artigas, F., 1997. Preferential potentiation of the effects of serotonin uptake inhibitors by 5-HT1A receptor antagonists in the dorsal raphe pathway: role of somatodendritic autoreceptors. J. Neurochem. 68, 2593–2603.

Schreiber, S., Backer, M.M., Yanai, J., Pick, C.G., 1996. The antinociceptive effect of fluvoxamine. Eur. Neuropsychopharmacol. 6, 281–284.

Schreiber, S., Bleich, A., Pick, C.G., 2002. Venlafaxine and mirtazapine: different mechanisms of antidepressant action, common opioid-mediated antinociceptive effects—a possible opioid involvement in severe depression? J. Mol. Neurosci. 18, 143–149.

Shapira, N.A., McConville, B.J., Pagnucco, M.L., Norman, A.B., Keck Jr., P.E., 1997. Novel use of tramadol hydrochloride in the treatment of Tourette's syndrome. J. Clin. Psychiatry 58, 174–175.

Shapira, N.A., Verduin, M.L., DeGraw, J.D., 2001. Treatment of refractory major depression with tramadol monotherapy. J. Clin. Psychiatry 62, 205–206.

Spencer, C., 2000. The efficacy of intramuscular tramadol as a rapid-onset antidepressant. Aust. N. Z. J. Psychiatry 34, 1032–1033.

Sprouse, J.S., Aghajanian, G.K., 1987. Electrophysiological responses of serotoninergic dorsal raphe neurons to 5-HT1A and 5-HT1B agonists. Synapse 1, 3–9.

Stamford, J.A., Davidson, C., McLaughlin, D.P., Hopwood, S.E., 2000. Control of dorsal raphe 5-HT function by multiple 5-HT(1) autoreceptors: parallel purposes or pointless plurality? Trends Neurosci. 23, 459–465.

Tatarczynska, E., Klodzinska, A., Stachowicz, K., Chojnacka-Wojcik, E., 2004. Effect of combined administration of 5-HT1A or 5-HT1B/1D receptor antagonists and antidepressants in the forced swimming test. Eur. J. Pharmacol. 487, 133–142.

Wang, Q.P., Nakai, Y., 1994. The dorsal raphe: an important nucleus in pain modulation. Brain Res. Bull. 34, 575–585.

Zimmermann, M., 1983. Ethical guidelines for investigations of experimental pain in conscious animals. Pain 16, 109–110.