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Tianeptine prevents respiratory depression without affecting analgesic effect of opiates in conscious rats

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ABSTRACT

Respiratory depression remains an important clinical problem that limits the use of opiate analgesia. Activation of AMPA glutamate receptors has been shown to reverse fentanyl-induced respiratory changes. Here, we explored whether tianeptine, a drug known for its ability to phosphorylate AMPA receptors, can be used to prevent opiate-induced respiratory depression. A model of respiratory depression in conscious rats was produced by administration of morphine (10 mg/kg, i.p.). Rats were pre-treated with test compounds or control solutions 5 min prior to administration of morphine. Respiratory activity was measured using whole-body plethysmography. In conscious animals, tianeptine (2 and 10 mg/kg, ip) and DP-201 (2-(4-((3-chloro-6-methyl-5,5-dioxido-6,11-dihydrodibenzo[c,f][1,2] thiazepin-11-yl)amino)butoxy)acetic acid; tianeptine analogue; 2 mg/kg, ip) triggered significant (~30%) increases in baseline respiratory activity and prevented morphine-induced respiratory depression. These effects were similar to those produced by an ampakine CX-546 (15 mg/kg, ip). The antinociceptive effect of morphine (hot plate test) was unaffected by tianeptine pre-treatment. In conclusion, the results of the experiments conducted in conscious rats demonstrate that systemic administration of tianeptine increases respiratory output and prevents morphine-induced respiratory depression without interfering with the antinociceptive effect of opiates.

1. Introduction

Opiates are the most commonly prescribed agents for the relief of postoperative pain. Of all their unwanted side effects, respiratory depression is of a greatest concern. The ability of opiates to reduce respiratory rate and tidal volume, as well as respiratory sensitivity to CO_2 has been long known (Shook et al., 1990). Opiate-induced respiratory depression is a life-threatening condition and a leading cause of death that may arise not only from overdose, but also during routine procedures supervised by clinicians, including surgical anaesthesia, post-operative analgesia, and as a result of routine out-patient management of pain from cancer, accidents, or other illnesses (Dahan et al., 2010).

Although only 0.5–1.2% of all adverse drug effects caused by prescription medications are respiratory in nature, these are serious and much more common in the context of patient controlled analgesia (Overdyk et al., 2007). Certain conditions predispose to respiratory depression such as morbid obesity, sleep apnoea, severe pain (patients receiving high doses of opiates) and chronic lung disease. Up to ~35% of all surgical patients fall into these high-risk categories.

Central respiratory drive is generated by bilaterally organised groups of respiratory neurones located in the ventrolateral regions of the medulla oblongata (Richter and Spyer, 2001; Feldman and Del Negro, 2006). Medullary respiratory network has been identified as being responsible for the decrease in the respiratory output following systemic administration of opiates (Ren et al., 2006). In addition, opiates suppress the activity of hypoglossal (XII) motoneurones which innervate tongue muscles and are vital for maintaining upper-airway patency. In emergency situations, administration of μ-opioid receptor antagonists such as naloxone is highly effective, but it also takes the patient out of analgesia. Therefore, development or identification of novel compounds that prevent respiratory depression associated with opiate analgesia without interfering with their

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analgesic effects is an important task.

The α-amino-3-hydroxy-5-methyl-4-isoxazole propionate (AMPA) glutamate receptors mediate excitatory inputs to XII motoneurones. Positive allosteric modulation of AMPA receptors by a group of compounds called AMPAkines (such as CX717 and others) has been shown to block as well as reverse respiratory depression induced by fentanyl in animal models and human volunteers (Oertel et al., 2010).

Tianeptine is a singular antidepressant that is proposed to be a "neuroplasticity enhancer" (Kole et al., 2002; McEwen et al., 2009; Szegedi et al., 2011). The exact underlying molecular mechanisms of its action remains unknown, but it has been shown that tianeptine increases the rate of phosphorylation at both the Ser831 and Ser845 residues of GluA1 subunit of AMPA receptors (Svenningsson et al., 2007; Qi et al., 2009; Barkóczi et al., 2012). There is evidence that tianeptine enhances the amplitude of the excitatory post-synaptic potentials (fEPSPs) in murine hippocampal slices – the effect which was blocked by kinase inhibitors (Zhang et al., 2013). High efficacy of AMPAkines in reducing opiate-induced respiratory depression suggested that the agents with a similar pharmacological profile may have a similar effect. Since tianeptine is capable of facilitating AMPA-mediated glutamatergic transmission we tested the effect of this compound on opiate-induced respiratory depression in an animal model.

2. Materials and methods

Experiments were performed in accordance with the European Commission Directive 86/609/EEC (European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes) and the UK Home Office (Scientific Procedures) Act (1986) with project approval from the respective Institutional Animal Care and Use Committees.

2.1. Experimental animals

A total of 82 male Sprague-Dawley rats (200–250 g body weight) were used in this study. Animals were fed *ad libitum* with a commercial rodent feed and had free access to drinking water. Animals were kept in automatically controlled environmental conditions set to maintain temperature at 22–24 °C with a relative humidity of 30–70%, and a 12:12 h light:dark cycle. At the end of the experiments the animals were humanely euthanized by an overdose of anaesthetic (pentobarbitone sodium, 200 mg/kg, ip).

2.2. Assessment of the respiratory activity

Whole-body plethysmography was used to measure respiratory rate (f_R, in breaths per minute), tidal volume (V_T, in microlitres per gram of body weight) and minute ventilation ($V_E = f_R \times V_T$) in conscious rats as described (Trapp et al., 2008, 2011). Briefly, the animals were placed in a recording chamber (~700 ml) flushed continuously with a humidified mixture of 79% nitrogen and 21% oxygen (temperature 22–24 °C). Level of CO_2 in the chamber was monitored online using a fast-response CO_2 analyser (Capstar 100, CWE). The animals were allowed ~40 min to acclimatize to the chamber environment (21% O_2, 79% N_2 and $< 0.3\%$ CO_2) before measurements of baseline ventilation were taken.

2.3. Model of opiate-induced respiratory depression

Respiratory depression in conscious rats was induced by intramuscular (10 mg/kg) administration of morphine (May and Baker). Prior to morphine administration, animals were randomly assigned to the experimental groups and received an intraperitoneal injection of either saline solution (control), tianeptine (2 mg/kg or 10 mg/kg, Kemprotec Ltd.), AMPAkine CX-546 (15 mg/kg, Tocris) or tianeptine analogue DP-201 (2 mg/kg, Peakdale Molecular Limited) (Table 1).

2.4. Assessment of the nociceptive threshold

Rats were injected with saline (ip) or tianeptine (10 mg/kg, ip) followed in 30 min by administration of morphine (10 mg/kg, im). Animals were then placed one at a time on a hot plate (52.5 °C) at intervals of 30, 90 and 240 min after morphine administration. Latency to respond to the heat stimulus was measured by the amount of time it took the animal to lick one of its rear paws or until cut-off time was reached (90 s).

2.5. Data acquisition and analysis

The data were acquired using Power1401 interface (CED Ltd., Cambridge, UK), saved and analysed off-line using *Spike2* software (CED). Data are presented as means ± SEM. Group data were compared by two-way ANOVA with repeated measures followed by Bonferroni's post-hoc analysis or one-way ANOVA followed by Tukey–Kramer's post-hoc analysis, as appropriate. Differences between the experimental groups with $P < 0.05$ were considered significant.

3. Results

3.1. The effect of tianeptine on the respiratory activity and opiate-induced respiratory depression in rats

In conscious rats, systemic administration of tianeptine (2 mg/kg, ip) increased the respiratory activity (by ~30%) 5 min after the

Table 1
Summary of the experimental groups.

Experiment	*n*	Test material	Anaesthesia	Route	Dose	Treatment
RD	20	Saline	Conscious	ip		Morphine, 10 mg/kg, im
RD	10	Tianeptine	Conscious	ip	2 mg/kg	Morphine, 10 mg/kg, im
RD	10	Tianeptine	Conscious	ip	10 mg/kg	Morphine, 10 mg/kg, im
RD	4	CX546	Conscious	ip	15 mg/kg	Morphine, 10 mg/kg, im
RD	6	DP-201	Conscious	ip	2 mg/kg	Morphine, 10 mg/kg, im
NT	5		Conscious			Saline, im
NT	5		Conscious			Morphine, 10 mg/kg, im
NT	5	Saline	Conscious	ip		Morphine, 10 mg/kg, im
NT	5	Tianeptine	Conscious	ip	10 mg/kg	Morphine, 10 mg/kg, im
NT	7	Saline	Conscious	ip		Morphine, 5 mg/kg, im
NT	5	Tianeptine	Conscious	ip	10 mg/kg	Morphine, 5 mg/kg, im

RD – Respiratory depression,
NT – Nociceptive threshold, ip – intraperitoneal, im – intramuscular.