ELSEVIER

European Journal of Pharmacology 333 (1997) 177–179

ejp

Effect of nociceptin on heart rate and blood pressure in anaesthetized rats

Sandro Giuliani *, Manuela Tramontana, Alessandro Lecci, Carlo Alberto Maggi

Pharmacology Department, Menarini Ricerche, Florence, Italy

Received 17 April 1997; accepted 1 July 1997

Abstract

We now report on the effects of nociceptin, the endogenous ligand for the orphan opioid-like ORL1 receptor, on cardiovascular parameters (blood pressure and heart rate) in urethane-anaesthetized rats. Nociceptin dose dependently (10–100 nmol/kg i.v.) produced a transient (< 10 min) hypotension and bradycardia. The reduction in blood pressure was inhibited by guanethidine pretreatment and unaffected by bilateral cervical vagotomy. The bradycardia elicited by nociceptin was reduced after bilateral vagotomy and by guanethidine and was abolished by the combination of the two treatments. These findings indicate that nociceptin exerts a pronounced depressant effect on cardiovascular function which is produced indirectly through a concomitant inhibition and activation, respectively, of the sympathetic and parasympathetic outflows to the cardiovascular system. © 1997 Elsevier Science B.V.

Keywords: Nociceptin; Blood pressure; Hypotension; Heart rate; Vagal bradycardia

1. Introduction

The novel neuropeptide, nociceptin, is considered the putative endogenous ligand for the opioid-like ORL1 receptor which is expressed in the central and peripheral nervous system (Wang et al., 1994). ORL1 transcripts have been identified in brain regions known to regulate blood pressure (Meunier et al., 1995) but their role in regulating cardiovascular function, is not yet settled. Recently, evidence has been presented indicating that nociceptin produces vasorelaxation of precontracted cat isolated arterial rings (Gumusel et al., 1997), suggesting a peripheral site of action for regulating vascular tone. Nociceptin also has a prejunctional site of action for inhibiting evoked transmitter release in the peripheral nervous system (Berzetei-Gurske et al., 1996; Giuliani and Maggi, 1996, 1997; Calò et al., 1996): a similar action may indirectly influence cardiovascular function. We have now investigated the activity of intravenous nociceptin on cardiovascular parameters (systemic blood pressure and heart rate) in anaesthetized rats.

2. Materials and methods

Male albino rats Wistar strain (Charles River, Calco, Italy), weighing 340–400 g, were anaesthetized with urethane (1.2 g/kg s.c.). The body temperature was kept constant at 36.5°C and the animals were tracheotomized.

The right jugular vein was cannulated for i.v. administration of drugs. Blood pressure was recorded through a polyethylene catheter inserted into the left carotid artery, connected to a pressure transducer and a HP 8805D pressure amplifier. The blood pressure signal was used to trigger a cardiotachometer (HP 15050A) for heart rate recording through a medium gain amplifier HP 8802A. Only one dose of nociceptin was administered to each animal and the effect of nociceptin was followed until the values returned to their resting levels.

In some experiments both vagi were sectioned at the cervical level (bilateral vagotomy). These animals were artificially ventilated by means of a Basile respirator for small rodents (70 strokes/min, 0.8 ml/100 g body weight).

Some experiments were performed in rats pretreated with guanethidine (68 μmol/kg s.c. administered in two equally divided doses, 14 and 1 h before the experiment).

All values in the text, tables or figures are means ± SEM. Statistical analysis was performed by means of Student's *t*-test for paired data or by means of one-way

* Corresponding author. Tel.: (39-55) 568-0350; Fax: (39-55) 568-0419.

Table 1
Resting cardiovascular parameters of urethane-anaesthetized rats, controls or following bilateral vagotomy or guanethidine pretreatment

	No.	Systolic blood pressure (mmHg)	Diastolic blood pressure (mmHg)	Heart rate (beats/min)
Control	19	145 ± 2	71 ± 3	405 ± 11
Vagotomy	5	137 ± 8	74 ± 5	430 ± 12
Guanethidine	6	131 ± 11	47 ± 3 *	361 ± 19
Vagotomy + guanethidine	5	159 ± 11	63 ± 2	416 ± 10

Each value is the mean ± SEM.
* $P < 0.05$, significantly different from control.

analysis of variance (ANOVA), followed by the Dunnett test for multiple comparisons, when applicable. A P level < 0.05 was considered statistically significant.

The drugs used were nociceptin (Tocris Cookson, Bristol, UK) and guanethidine sulfate (Sigma, St. Louis, MO, USA).

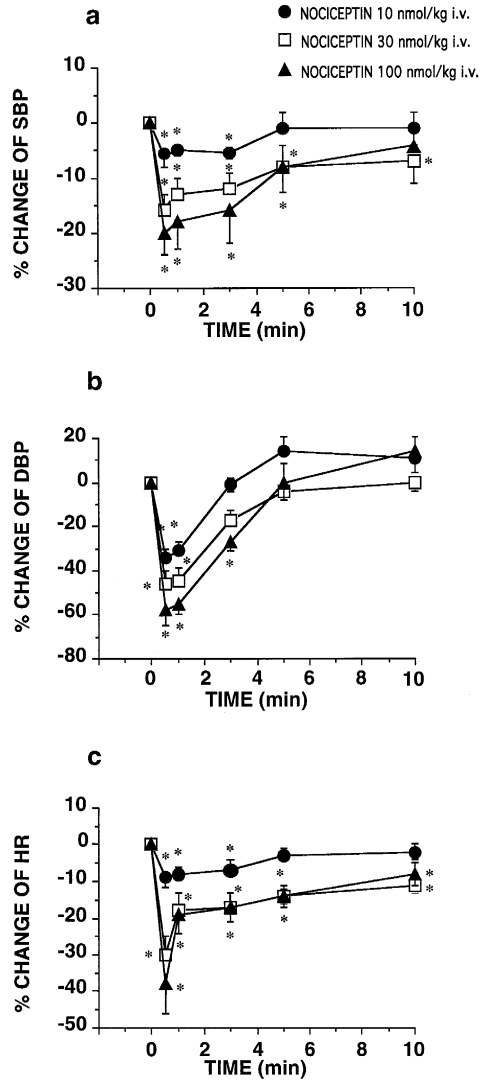

Fig. 1. Time-course of the effect of intravenous nociceptin (● 10 nmol/kg, □ 30 nmol/kg, ▲ 100 nmol/kg) on systolic blood pressure (SBP, panel a), diastolic blood pressure (DBP, panel b) and heart rate (HR, panel c) in urethane-anaesthetized rats. Each values is the mean ± SEM of 6-7 experiments. * $P < 0.05$, significantly different from baseline values.

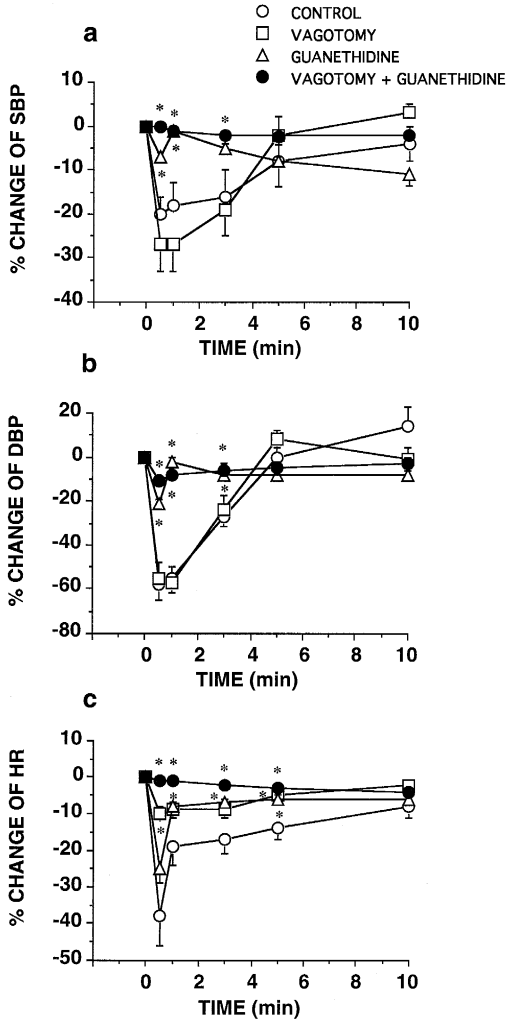

Fig. 2. Effect of i.v. nociceptin (100 nmol/kg) on systolic blood pressure (SBP, panel a), diastolic blood pressure (DBP, panel b) and heart rate (HR, panel c) in urethane-anaesthetized rats in control animals (○), after bilateral cervical vagotomy (□), guanethidine pretreatment (△) and combination of both (●). Each values is the mean ± SEM of 5-7 experiments. * $P < 0.05$, significantly different from controls.

3. Results

The resting values for blood pressure and heart rate in the various experimental groups are shown in Table 1. Nociceptin produced a dose-dependent (10–100 nmol/kg) transient hypotension and bradycardia. These responses reached a maximum within 15–30 s and persisted for 3–10 min (Fig. 1). At a dose of 100 nmol/kg the peak effects were: -26 ± 7 mmHg, -42 ± 5 mmHg and -155 ± 32 beats/min ($n = 7$) for systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR), respectively. This dose was selected for evaluating the role of the vagi and of the sympathetic nervous system in the cardiovascular effects of nociceptin. The inhibitory effect of nociceptin on both SBP and DBP was reduced by guanethidine (68 μmol/kg s.c.) pretreatment (65 ± 4 and $76 \pm 2\%$ inhibition, $n = 6$) but unaffected by bilateral vagotomy alone. The combination of both provoked a greater reduction of the nociceptin-induced hypotensive effects on SBP and DBP (100 ± 1 and $83 \pm 1\%$ inhibition, $n = 5$, respectively) (Fig. 2).

The bradycardia elicited by nociceptin was markedly ($72 \pm 5\%$) reduced by bilateral vagotomy and to a smaller extent ($42 \pm 8\%$) by guanethidine and was abolished by the combination of both ($n = 5$) (Fig. 2).

4. Discussion

The present findings indicate that nociceptin exerts a profound depressant action on cardiovascular function in anaesthetized rats: all the effects of nociceptin were inhibited if bilateral cervical vagotomy and guanethidine pretreatment were combined, indicating that these effects are indirectly mediated via the autonomic nervous system. Therefore, the recently described vasorelaxant action of nociceptin (Gumusel et al., 1997) is unlikely to be involved in the hypotensive effect described in this study.

The intense bradycardia provoked by nociceptin was largely reduced by vagotomy, implying that activation of the parasympathetic outflow to the heart was involved: indeed atropine pretreatment (5 μmol/kg i.v., $n = 2$) reduced the nociceptin-induced bradycardia by about 50 %, indicating that acetylcholine release was the main effector mechanism involved (data not shown). It may be speculated that, owing to the marked reduction in heart rate, the concomitant hypotension was secondary to a reduced cardiac pump function. However, the hypotension was little affected by vagotomy alone, implying that a second mechanism is required to account for the entire spectrum of nociceptin cardiovascular effects. The hypotensive response was largely prevented by guanethidine pretreatment. This indicates that an inhibitory influence of nociceptin on the sympathetic outflow to the cardiovascular system is mainly responsible for the observed hypotension. In addition, guanethidine pretreatment partly reduced the nociceptin-induced bradycardia and the combined treatment (vagotomy plus guanethidine) was required to totally eliminate the bradycardic response to nociceptin. Thus, there must be, in addition to activation of the parasympathetic outflow to the heart, a concomitant inhibition of the sympathetic drive. It should be noted that both the parasympathetic and the sympathetic outflows to the heart are tonically active in the regulation of the heart rate of urethane-anaesthetized rats, since the administration of atropine or propranolol induces tachycardia and bradycardia, respectively, in these animals (Maggi and Meli, 1986).

In conclusion, the present findings demonstrate that nociceptin exerts a profound depressant action on cardiovascular function which is ascribable to a concomitant activation of the vagal outflow to the heart and inhibition of sympathetic cardiovascular tone.

References

Berzetei-Gurske, I.P., Schwartz, R.W., Toll, L., 1996. Determination of activity for nociceptin in the mouse vas deferens. Eur. J. Pharmacol. 302, R1–R2.

Calò, G., Rizzi, A., Bogoni, G., Neugebauer, V., Salvadori, S., Guerrini, R., Bianchi, C., Regoli, D., 1996. The mouse vas deferens: A pharmacological preparation sensitive to nociceptin. Eur. J. Pharmacol. 311, R3–R5.

Giuliani, S., Maggi, C.A., 1996. Inhibition of tachykinin release from peripheral endings of sensory nerves by nociceptin, a novel opioid peptide. Br. J. Pharmacol. 118, 1567–1569.

Giuliani, S., Maggi, C.A., 1997. Prejunctional modulation by nociception of nrve-mediated inotropic responses in guinea-pig left atrium. Eur. J. Pharmacol. 332, 231–236.

Gumusel, B., Hao, Q., Hyman, A., Chang, J.K., Kapusta, D.R., Lippton, H., 1997. Nociceptin: an endogenous agonist for central opioid like 1 (ORL1) receptors possesses systemic vasorelaxant properties. Life Sci. 60, PL141–PL145.

Maggi, C.A., Meli, A., 1986. Suitability of urethane anaesthesia for physiopharmacological investigations in various systems. Part 2. Cardiovascular system. Experientia 42, 292–297.

Meunier, J.C., Mollereau, C., Toll, L., Suadeau, C., Moisand, C., Alvinerie, P., Butour, J.L., Guillemot, J.C., Ferrara, P., Monsarrat, B., Mazarguil, H., Vassart, G., Parmentier, M., Costentin, J., 1995. Isolation and structure of the endogenous agonist of opioid receptor-like ORL1 receptor. Nature 377, 532–535.

Wang, J.B., Johnson, P.S., Imai, Y., Persico, A.M., Ozenberger, B.A., Eppler, C.M., Uhl, G.R., 1994. cDNA cloning of an orphan receptor gene family member and its splice variant. FEBS Lett. 348, 75–79.