European Journal of Pharmacology 699 (2013) 200–206

Contents lists available at SciVerse ScienceDirect

European Journal of Pharmacology

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

Neuropharmacology and analgesia

Retrodialysis of N/OFQ into the nucleus accumbens shell blocks cocaine-induced increases in extracellular dopamine and locomotor activity

Jacqueline Vazquez-DeRose [a], Gregory Stauber [a], Taline V. Khroyan [a], Xinmin Simon Xie [b], Nurulain T. Zaveri [c], Lawrence Toll [d,*]

[a] SRI International, Menlo Park, CA 94025, USA
[b] AfaSci Research Laboratory, Redwood City, CA 94063, USA
[c] Astraea Therapeutics, Mountain View, CA 94043, USA
[d] Torrey Pines Institute for Molecular Studies, 11350 Staj Village Porkway, Port St. Lucie, FL 34987, USA

ARTICLE INFO

Article history:
Received 13 June 2012
Received in revised form
21 November 2012
Accepted 22 November 2012
Available online 5 December 2012

Keywords:
Microdialysis
Nociceptin
N/OFQ
Nucleus accumbens
NOP receptor
Cocaine
Rat
Locomotion

ABSTRACT

Nociceptin (N/OFQ) has been implicated in a variety of neurological disorders, most notably in reward processes and drug abuse. N/OFQ suppresses extracellular dopamine in the nucleus accumbens (NAc) after intracerebroventricular injection. This study sought to examine the effects of retrodialyzed N/OFQ on the cocaine-induced increase in extracellular dopamine levels in the NAc, as well as locomotor activity, in freely moving rats. 1.0 μM, 10 μM, and 1 mM N/OFQ, in the NAc shell, significantly suppressed the cocaine-induced dopamine increase in the NAc, while N/OFQ alone had no significant effect on dopamine levels. Co-delivery of the selective NOP receptor antagonist SB612111 ([(−)-cis-1-Methyl-7-[[4-(2,6-dichlorophenyl)piperidin-1-yl]methyl]-6,7,8,9-tetrahydro-5 H-benzocyclohepten-5-ol] reversed the N/OFQ suppression of cocaine-induced dopamine in the NAc, suggesting that this is an NOP receptor-mediated effect. Using a novel system to assess locomotion, we measured various motor activities of the animals with simultaneous microdialysis from the home cage. Cocaine produced an expected increase in total activity, including horizontal movement and rearing behavior. Retrodialysis of N/OFQ with cocaine administration affected all motor activities, initially showing no effect on behavior, but over time inhibiting cocaine-induced motor behaviors. These results suggest that N/OFQ can act directly in the NAc shell to block cocaine-induced increases in extracellular dopamine levels. Extracellular dopamine and locomotor activity can be dissociated within the NAc and may reflect motor output differences in shell versus core regions of the NAc. These studies confirm the widespread involvement of NOP receptors in drug addiction and further validate the utility of an NOP receptor agonist as a medication for treatment of drug addiction.

1. Introduction

The endogenous neuropeptide, Nociceptin/Orphanin FQ (N/OFQ) (Meunier et al. 1995; Reinscheid et al. 1995), and its cognate receptor, NOP, are found throughout the brain and are involved in a large number of neurological processes including anxiety, memory, feeding, pain, and reward, among others. There is a moderate to high density of NOP receptors in areas implicated in drug abuse and reward, including the nucleus accumbens (NAc), ventral tegmental area (VTA), medial prefrontal cortex, lateral hypothalamus, amygdala, and the bed nucleus of stria terminalis (Neal et al., 1999a, 1999b). N/OFQ injected intracerebroventricularly (i.c.v.) attenuates the rewarding properties of several common drugs of abuse. Specifically, N/OFQ blocks conditioned place preference (CPP) induced by morphine, cocaine, amphetamines, and alcohol (Ciccocioppo et al., 2000; Kotlinska et al., 2003b; Murphy et al., 1999; Sakoori and Murphy, 2004; Zhao et al., 2003), alcohol self-administration, and stress-induced reinstatement of alcohol self-administration (Ciccocioppo et al., 2004; Martin-Fardon et al., 2000). N/OFQ also blocks the stimulant effects associated with drugs of abuse. For example, i.c.v. injections of N/OFQ attenuate the acute motor stimulatory effects of cocaine (Lutfy et al., 2001; Narayanan et al., 2004).

Microdialysis studies, measuring extracellular dopamine following N/OFQ administration, have complemented the behavioral findings discussed above. N/OFQ decreases basal dopamine levels and attenuates morphine and cocaine-induced increases of dopamine levels in the NAc (Di Giannuario et al., 1999; Lutfy et al., 2001; Murphy et al.,

* Corresponding author. Tel.: +1 772 345 4714; fax: +1 772 345 3649.
E-mail addresses: jacqueline.vazquez@sri.com (J. Vazquez-DeRose), gregory.stauber@sri.com (G. Stauber), taline.khroyan@sri.com (T.V. Khroyan), simonxie@afasci.com (X.S. Xie), nurulain@astraeatherapeutics.com (N.T. Zaveri), ltoll@tpims.org (L. Toll).

0014-2999/$ - see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.ejphar.2012.11.050

1996). Furthermore, an injection of N/OFQ (1 mM) directly into the VTA, is sufficient to attenuate cocaine-induced increases in NAc dopamine and locomotor activity (Murphy and Maidment, 1999). However, it should be noted that striatal administration of high concentrations of N/OFQ can have opioid-like activity, inducing a naloxone reversible increase in dopamine levels in the nucleus accumbens (NAc) (Konya et al., 1998).

To address whether the NOP receptor system can directly influence dopamine release in the NAc, the effect of retrodialyzed N/OFQ in the NAc shell on the cocaine-induced increases in extracellular dopamine levels, in this region, was examined. In parallel we also examined whether intra-NAc N/OFQ could alter cocaine-induced activity, using the novel SmartCage™ apparatus, under identical experimental conditions. Results indicated that 1 μM–1000 μM N/OFQ dialyzed within the NAc shell blocked the cocaine-induced increase in extracellular dopamine in this brain region. Additionally, intra-NAc N/OFQ administration also induced a delayed decrease in cocaine-induced hyperactivity.

2. Materials and methods

2.1. Animals

Male Sprague-Dawley rats (200–250 g; Harlan Laboratories, Hayward, CA) were housed under constant ambient conditions on a 12 h light/dark cycle (lights on at 06:00 h) with food and water ad libitum. All animals were handled and habituated to the microdialysis testing procedures for 1 week prior to surgery and 1 week post-surgery. All studies were conducted in accordance with the Guide for the Care and Use of Laboratory Animals (National Academy of Sciences Press, Washington, DC, 1996) and were approved by the Institutional Animal Care and Use Committee at SRI International. Every effort was made to minimize animal discomfort throughout the experimental protocols.

2.2. Drugs

N/OFQ was obtained from the NIDA drug supply program. The NOP receptor antagonist SB612111 ([(−)-cis-1-Methyl-7-[[4-(2,6-dichlorophenyl)piperidin-1-yl]methyl]-6,7,8,9-tetrahydro-5H-benzocyclohepten-5-ol] was synthesized at SRI International using previously reported methodology (Barlocco et al., 2006). Drugs that were delivered by reverse dialysis were dissolved in artificial cerebrospinal fluid (aCSF, 124 mM NaCl, 2.2 mM KCl, 1.3 mM KH_2PO4, 1.3 mM $MgSO_4$, 20 mM $NaHCO_3$, and 2.0 mM $CaCl_2$; pH 6.0–6.2). Cocaine (20 mg/kg, Sigma, St. Louis, MO) was prepared in 0.9% saline and administered intraperitoneally (i.p.) to the animals. All drug solutions were made on the day of each experiment and serially diluted to their final concentrations. The 1 μM, 10 μM, and 1000 μM concentrations of N/OFQ and the 100 μM of SB612111 (NOP antagonist) were delivered by reverse dialysis.

2.3. Surgery

The animals were anesthetized with isoflurane (2–3%) and their core temperature maintained at 37 °C using a thermoregulated heating pad in conjunction with a rectal probe. The rats were implanted with microdialysis guide cannulae (CMA/12; CMA Microdialysis, Chelmsford, MA), which were stereotaxically implanted 2 mm above the NAc (shell) [AP,+1.7; L, +0.8; DV,−6.0 as calculated relative to bregma and skull surface, according to Paxinos and Watson (Paxinos and Watson, 1997). The cannulae were affixed to the head with two small screws and dental cement. All animals were allowed at least 1 week of recovery and several hours (4–6 h) of daily habituation to the microdialysis environment prior to the initiation of experiments.

2.4. Assessment of dopamine levels using microdialysis and high performance liquid chromatography (HPLC) with electrochemical detection (EC)

Concentric microdialysis probes with 2 mm exposed membrane (0.5 mm diameter, 20 kDa cutoff; CMA 12, CMA Microdialysis) were inserted into the microdialysis cannula 18 h prior to sample collection, to allow for dopamine stabilization, and perfused with aCSF at a rate of 1 μl/min. Overnight perfusate was discarded. On the morning of each microdialysis session, samples were collected at 20 min intervals into vials containing 5 μL of 1 mM oxalic acid (to prevent degradation of monoamine transmitters) and kept at 4 °C for the duration of the experiment. The samples were subsequently frozen at −70 °C and later analyzed in several experiment batches using HPLC/EC (ESA-Dionex, Inc. Sunnyvale, CA).

Microdialysis samples were then injected into an HPLC system via an ESA model 542 autosampler and separated on an ESA MD-150/RP-C18 analytical column (150 × 3.2 mm, 3 μm), perfused with ESA mobile phase MD-TM type II (#70-5049 P, Chelmsford, MA) at a flow rate of 0.6 mL/min. Dopamine was detected by oxidation using an ESA CouloChem III detector equipped with an ESA 5020 guard cell (+300 mV) and an ESA 5014b dual electrode analytical cell (E1, −100 mV; E2, +200 mV). Chromatographic data were acquired and analyzed using EZChrome Elite software. The HPLC system was calibrated at the start of each set of experimental batch of samples, using external dopamine standards.

2.5. Assessment of motor behavior using SmartCage™

In a separate series of experiments, motor behavior in freely moving animals was assessed using the SmartCage™ technology (AfaSci, Inc., Redwood City, CA) (Khroyan et al., 2012; Xie et al., 2011). SmartCage™ uses a USB-cable linked, noninvasive rodent behavior monitoring system in conjunction with the animal's home cage. Microdialysis balance arms and dual channel swivels (CMA Microdialysis) were adapted to a home cage, with the SmartCage™ apparatus positioned around the home cage, so that the animals could remain in their home environment while motor activity could be measured. Motor traces were generated to represent 20 min of activity/interval over time in the animal's home cage from left to right. Motor behavior was quantified as percent active time, locomotion (cm), and the incidence of rearing. Calculation of locomotion was based on breakage of the lower infra-red beams of the SmartCage™, and rearing was defined as the breakage of the upper infra-red beams. Percent active time encompassed the time that the animal was moving across the cage (i.e. breaking consecutive horizontal beams), rearing, and also more refined movements including sniffing and grooming behavior.

2.6. Experimental design

In the first series of microdialysis experiments animals received cocaine alone (20 mg/kg, i.p.; $N=8$), N/OFQ alone (1000 μM, by reverse dialysis; $N=7$), or cocaine (20 mg/kg, i.p.) with N/OFQ (reverse dialysis; 1.0 μM, 10 μM, or 1000 μM; $N=$ 6–11/group). The dose of cocaine was chosen to be similar to other microdialysis studies (Chefer et al., 2003). Initially the 1000 μM dose of N/OFQ was chosen since it was the concentration used previously in the literature (Murphy and Maidment,