Pharmacology, Biochemistry and Behavior 110 (2013) 112–116

Contents lists available at ScienceDirect

Pharmacology, Biochemistry and Behavior

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

Oxycodone-induced conditioned place preference and sensitization of locomotor activity in adolescent and adult mice

Keiichi Niikura *, Ann Ho, Mary Jeanne Kreek, Yong Zhang

Laboratory of the Biology of Addictive Diseases, The Rockefeller University, 1230 York Avenue, New York, NY 10065, USA

ARTICLE INFO

Article history:
Received 11 February 2013
Received in revised form 25 May 2013
Accepted 22 June 2013
Available online 1 July 2013

Keywords:
Adolescent
Conditioned place preference
Locomotor sensitization
Oxycodone

ABSTRACT

Nonmedical use of the prescription opioid oxycodone has become a major public health problem in the United States, with special concern for adolescents. Although adults and adolescents have different sensitivities for drugs, little is known about the rewarding effects of oxycodone in adolescents compared to adults, even in rodent models. Here, we investigate sensitivity to oxycodone by the conditioned place preference assay of conditioned reward, and effect on the locomotor activity in adolescent (4 weeks old) and adult (10 weeks old) C57BL/6J mice. Mice of both ages were trained with multiple doses of oxycodone (0, 0.3, 1, and 3 mg/kg) and showed conditioned preference in a dose-dependent manner. The adult mice developed conditioned preference to the lowest dose tested (0.3 mg/kg), but adolescent mice did not. Dose-dependent oxycodone-induced increases in locomotor activity were observed across the conditioning session. Interestingly, adolescent mice developed greater sensitization to the locomotor-activating effects of oxycodone than adult mice. Thus differences in sensitivity to oxycodone, such as the lower initial sensitivity for conditioned preference but greater locomotor sensitization in adolescent mice, may indicate contributing factors in oxycodone abuse and later addiction in human adolescents.

1. Introduction

Although prescription opioid analgesics, such as oxycodone, are among the most effective medications for pain, they are also associated with the public health problem of illicit use of prescription opioids in the United States. Oxycodone is a mu-opioid agonist (MOP-r) and has been used for over half century in the United States. Compared with the prototypical MOP-r agonist morphine (also a major metabolite of heroin), potency of oxycodone is approximately twice as high, by the oral route (Benziger et al., 1997). Like morphine, oxycodone has rewarding effects (Rutten et al., 2011). However, unlike morphine, oxycodone is metabolized by Cytochrome P450 (CYP2D6 and CYP3A4) (see review Smith, 2009). Previous studies show that morphine causes MOP-r desensitization but poor internalization, compared to other MOP-r ligands (Keith et al., 1998; Whistler et al., 1999; Haberstock-Debic et al., 2003). The profile of oxycodone in this regard may differ from that of morphine, depending on in vitro conditions (Arttamangkul et al., 2008; Imai et al., 2011).

The adolescent brain is particularly susceptible to the effects of drugs of abuse. The brain circuits undergo developmental alterations during adolescence. For example, the number of dopamine receptors in the nigrostriatal and mesolimbic dopaminergic systems steadily increased after birth and peaks at 40 days of age in adolescent mice, then decreases at 60–80 days of age (Teicher et al., 1995). Mu-opioid receptor-stimulating [35S] GTPgammaS binding assessing mu-opioid receptor function showed a 20-fold increase between postnatal day five and adulthood (Talbot et al., 2005). Further, many studies have reported that adolescent rodents respond differently than adults to a number of drugs which act on the dopamine systems. For example, adolescents showed a reduced responsiveness to the locomotor effects of the psychostimulants amphetamine and cocaine (Spear and Brick, 1979; Bolanos et al., 1998). Conversely, these animals were more sensitive to the dopamine antagonist haloperidol (Spear et al., 1980) and exhibited an exaggerated locomotor response to morphine compared to that of adults (Spear et al., 1982).

We previously reported the effects of oxycodone self-administration and subsequent effects of oxycodone administration on striatal dopamine levels in adolescent and adult mice (Zhang et al., 2009b). We found that adult mice self-administered significantly more oxycodone than did adolescent mice during the acquisition of self-administration behavior. However, when examined later, in response to the lowest dose of experimenter-administered oxycodone tested, there was a significant increase in striatal dopamine levels in mice that had self-administered oxycodone during adolescence, but not in mice that had self-administered oxycodone as adults (Zhang et al., 2009b). In addition to our result, it has been reported that plasma ACTH response to CRH was enhanced in late adolescent male rats as a result of prenatal oxycodone exposure, but had no effect in females (Sithisarn et al., 2008). Taken together, these results suggest that oxycodone-induced rewarding effects differ between adolescent and adult mice. We hypothesize that adolescent and adult mice show different responses to the rewarding and locomotor stimulating effects of oxycodone, in which exposure to oxycodone is equalized across adolescents and adults. To test this

* Corresponding author. Tel.: +1 212 327 8490; fax: +1 212 327 8574.
E-mail address: kniikura@mail.rockefeller.edu (K. Niikura).

0091-3057/$ – see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.pbb.2013.06.010

hypothesis, the current study was carried out to compare oxycodone-induced conditioned rewarding effects and locomotor activity in both adolescent and adult C57BL/6J mice using a conditioned place preference paradigm. The locomotor stimulant effects may be useful markers for the addictive properties of drugs, due to a high homology between the brain regions underlying stimulant and rewarding effects of drugs (Wise and Bozarth, 1987). Sensitization of certain components of drug action has also been postulated as part of addiction mechanisms (Robinson and Berridge, 1993, 2001, 2003; Vanderschuren and Pierce, 2010).

2. Materials and methods

2.1. Animals

Male adolescent and adult (4 or 10 weeks old on arrival) C57BL/6J mice (Jackson Laboratory, Bar Harbor, ME) were housed in groups of four with free access to food and water in a light (12:12 h light/dark cycle, lights on at 0700 hours) and temperature (25 °C) controlled room. Animal care and experimental procedures were conducted according to the Guide for the Care and Use of Laboratory Animals (Institute of Laboratory Animal Resources Commission on Life Sciences National Research Council, 1996). The experimental protocols used were approved by the Institutional Animal Care and Use.

Committee of the Rockefeller University. See Table 1 for a description of the experimental time line and age of the mice.

2.2. Mouse place preference chambers

The mouse place preference chambers (model ENV-3013) were purchased from Med Associates (Med Associates, St. Albans, VT). Each chamber has three distinct compartments that can be separated by removable doors. Automated data collection is accomplished by individual infrared photobeams on a photobeam strip, with six beams in the white and black compartments and two beams in the smaller central gray compartment. The black compartment is 16.8 × 12.7 × 12.7 cm with a stainless-steel grid rod floor. The white compartment (also 16.8 × 12.7 × 12.7 cm) has a stainless-steel mesh floor (Zhang et al., 2002, 2012).

2.3. Locomotor activity and conditioned place preference determination

Four groups of seven mice in each age groups were studied, one group at each dose: 0, 0.3, 1, and 3 mg/kg of oxycodone. Valuable strain comparisons have been generated for setting up the schedule and opioid ligand dosage for CPP (e.g., Orsini et al., 2005). However, since we focus on the effect of oxycodone in different ages in the current study, the dosage and experimental paradigm are used based on previous studies within one strain. The doses chosen in the study were based on published data in adult mice (Narita et al., 2008). The experimental paradigm for the CPP study was based on our earlier studies (Schlussman et al., 2008; Zhang et al., 2009a). Experiments were performed in a dimly lit, sound-attenuated chamber described above. The study used an unbiased, counterbalanced design in which seven mice were randomly assigned to either the oxycodone or saline compartment on the first day. Half the animals had white and half had black as the oxycodone-paired side. During the preconditioning session, each animal was placed in the center compartment with free access to the black and white compartments, and the amount of time spent in each compartment was recorded for 30 min. During the conditioning sessions, mice were placed into and restricted to the appropriate compartment for 30 min after oxycodone or saline injection. Locomotor activity was assessed as the number of "crossovers," defined as breaking the beams at either end of the conditioning compartment. The animals were injected with oxycodone and saline on alternate days, for a total of eight conditioning sessions with four oxycodone and four saline trials for each animal. Conditioning sessions were conducted daily. The postconditioning test session (without injection; i.e., a drug-free state) was performed on the day after the last conditioning session, and was identical to the preconditioning session: Each mouse had free access to both white and black compartments. The difference between the pre- and postconditioning sessions in the amount of time spent on the drug-paired compartment was used to determine whether the mice had developed a conditioned place preference to oxycodone.

2.4. Statistical analysis

Analyses of variance (ANOVAs) of CPP, locomotor activity, and locomotor sensitization, followed by Newman–Keuls post hoc tests were used to examine the significance of differences in behavior between drug doses, ages and sessions. The results of each age group were also separately examined by ANOVA.

3. Results

A preliminary analysis was made to compare the amount of time spent during the preconditioning session in the compartment that was later paired with oxycodone to the one paired with saline. Two-way ANOVAs (Age × Side) showed no main effect of age or side, nor as there a significant interaction (means ± S.E.M. are shown in Table 2). Also, in the postconditioning test session, mice of both ages showed no increased amount of time spent on the oxycodone-paired compartment, regardless of which compartment was used for pairing.

3.1. Conditioned place preference in adolescent and adult mice

The increase in the amount of time spent on the drug-associated side, indicating a conditioned place preference, is shown in Fig. 1 in each age group of mice at each of four doses. Two-way ANOVA (Dose × Age) showed a significant main effect of Dose [$F(3,48) = 14.57$, $p < 0.0001$], with no significant difference between Age groups and no significant Dose × Age interaction. Of interest, Newman–Keuls post hoc tests showed that there was a significant difference in preference between the two age groups in response to the lowest dose (0.3 mg/kg) of oxycodone: adolescent mice did not show preference, whereas the adult mice did ($p < 0.05$). When the two age groups were examined separately by one-way ANOVA, adolescent mice showed a significant main effect of Dose [$F(3,24) = 9.631$, $p < 0.0002$]. A significant preference for the drug-paired side was not produced by the lowest dose, 0.3 mg/kg, but there was a significant preference induced by doses of 1 and 3 mg/kg compared to the saline control ($p < 0.05$ for 1 mg/kg, $p < 0.001$ for 3 mg/kg). In adult mice, there was a significant main effect of Dose on increased time on the drug-associated side [$F(3,24) = 7.314$, $p < 0.0012$],

Table 1
Time line: age in experiment.

	On arrival	Preconditioning	Conditioning	Postconditioning
Adolescent postnatal day	28	35	36–43	44
Adult postnatal day	70	77	78–85	86

Table 2
Amount of time spent during the preconditioning session in the compartment (sec).

	Saline side	Oxycodone side
Adolescent	652 ± 27	618 ± 19
Adult	666 ± 36	630 ± 27