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HIV-gp120 and physical dependence to buprenorphine

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Abstract

Background—Opioids are among the most effective and commonly used analgesics in clinical practice for severe pain. However, the use of opioid medications is clinically limited by several adverse properties including dependence. While opioid dependence is a complex health condition, the treatment of HIV-infected individuals with opioid dependence presents additional challenges. The goal of this study was to examine the physical dependence to buprenorphine in the context of HIV.

Methods—Young adult male rats (Sprague-Dawley) were pretreated with HIV-1 envelope glycoprotein 120 (gp120) injected into the periaqueductal grey area (PAG) and we examined the impact on physical dependence to opioid.

RESULTS—It was found that the physical dependence to methadone occurred earlier than that to buprenorphine, and that gp120 did not enhance or precipitate the buprenorphine withdrawal.

CONCLUSION—The results suggest that buprenorphine could be the better therapeutic option to manage opioid dependence in HIV.

Keywords

Buprenorphine; methadone; HIV-1; gp120 and physical dependence

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Contributors

K. Benamar designed the study and wrote the manuscript.

J. Palma, conducted the experiments

ME Abood revised the draft.

All the authors have contributed to and approved the final version of the manuscript.

Conflict of Interest

All authors declare that they have no conflicts of interest.

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1. INTRODUCTION

Buprenorphine is a semi-synthetic derivative of thebaine. It has a molecular weight of 467 and its structure is typically opioid with the inclusion of a C-7 side-chain containing a t-butyl group. Comparison of the antinociceptive effects of methadone and buprenorphine shows that 3 mg/kg for methadone is the effective analgesic dose, and 0.3 mg/kg for buprenorphine in rats using the hot-plate test (Bulka et al., 2004). Respiratory depression caused by opioids can be potentially life-threatening but is much less of a problem with buprenorphine than with many other opioids including morphine, hydromorphone, methadone, oxycodone, and transdermal fentanyl (Dahan et al., 2005). This advantage is due to the unique pharmacological characteristics of buprenorphine as a partial μ -agonist. Its ceiling effect, associated with a bell-shaped dose-response (D-R) curve with regard to respiratory depression, means that the risk to induce respiratory arrest does not linearly follow dose-increments of the drug. Although methadone is used as a pharmacotherapy for opioid dependence in HIV-1 infected individuals, its use has been associated with several adverse drug interactions with HIV-1 therapies that can produce either elevated methadone concentrations with toxicity, or decreased methadone levels with withdrawal (Gruber and McCance-Katz, 2010; McCance-Katz, 2005). In contrast, buprenorphine has not been shown to produce significant adverse drug interactions with antiretroviral therapy drugs such as delavirdine, efavirenz, nelfinavir, ritonavir or lopinavir/ritonavir (Gruber and McCance-Katz, 2010; McCance-Katz, 2005). The lack of drug interaction between antiretroviral therapy and buprenorphine is potentially an important advantage of buprenorphine treatment of opioid dependence in HIV-1 infected patients.

Opioid abuse and therapeutic use are frequently associated with HIV infection. With the global HIV prevalence estimated at 35.3 million, approximately 30% of HIV-positive individuals, within developed countries, are intravenous (IV) drug users, and it is the third most frequently reported risk factor for HIV-1 infection in the United States (CDC, 2009). In addition, pain is part of the clinical picture associated with HIV and AIDS. Opioids are among the most effective and commonly used analgesics in clinical practice for severe pain. However, the use of opioid medications is clinically limited by several adverse properties including dependence. Currently, there is no data available examining the physical dependence to buprenorphine in neuroAIDS. Therefore, we determined the physical dependence to these opioids in the context of HIV.

2. MATERIALS AND METHODS

2.1 Animals

All animal procedures were conducted in strict accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals and were approved by the Institutional Animal Care and Use Committee (IACUC). Male Sprague-Dawley rats (250–300 g; Harlan, Indianapolis, IN) were housed two per cage for at least one week before surgery and were fed laboratory chow and water *ad libitum* at ambient temperature of 21 ± 0.3 °C, and a 12 h light/12 h dark cycle.

2.2 Surgery procedures

Rats were anesthetized with an intraperitoneal injection of a mixture of ketamine hydrochloride and acepromazine maleate. For cannula implantation, each animal was placed in a stereotaxic instrument. A sterilized stainless steel C313G cannula guide (Plastics One Inc., Roanoke, VA) was implanted into the ventral and ventro-lateral PAG (Palma et al., 2011). The stereotaxic coordinates were as follows: 7.8 mm anterior to bregma, 0.5 mm from midline and 4 mm ventral to the dura mater (Paxinos and Watson, 2007).

2.3 Microinjection

The animals were habituated to the handling procedure necessary for microinjections during the recovery period. After the recovery period, rats were allowed to habituate to test chambers for 1 h before testing. The rats were gently restrained while the dummy stylets were removed and replaced with a C313I injector cannula (Plastics One Inc., Roanoke). Either vehicle or drug was microinjected into the PAG in a volume of 0.5 μ l. The C313I injector cannula was connected by polyethylene tubing to a 10- μ l Hamilton syringe. A volume of 0.5 μ l of drug or vehicle was delivered at a rate of 0.5 μ l per min and the internal cannula left in place an additional 90 sec to allow diffusion. Immediately thereafter, a dummy cannula (C313DC) was inserted into the cannula guide to prevent any contamination (Palma et al., 2011).

2.4 Drugs

Buprenorphine and methadone were obtained from the National Institute on Drug Abuse and dissolved in sterile, pyrogen-free saline. HIV-1 envelope glycoprotein gp120 recombinant viral protein was obtained from Advanced Biotechnologies (Columbia, MD).

2.5 Dependence and Withdrawal

The development of physical dependence was induced by repeated administrations of opioids. Rats were injected subcutaneously (s.c.) with buprenorphine or methadone at 9:00 a.m. daily. The rats were placed individually into test chambers consisting of boxes (50×35×45 cm) and allowed to acclimate for 30 min. Qualitative and/or quantitative evaluation of abstinence consists of measuring a series of withdrawal phenomena (Baldino et al., 1979; Cowan, 1981; Huang et al., 2009) after opioid discontinuation. Examples of behaviors observed are “wet-dog” shakes, teeth chattering, jumping, diarrhea, and flat posture. Simultaneously body weight was monitored.

2.6 Nociceptive test

A 52°C hot-plate (Ugo Basile, Varese, Italy) was used to measure the nociceptive response. The baseline response latency was obtained for each animal after two conditioning runs. Each rat was retested on the hot plate at 15 min and thereafter at 15-min intervals by using either jumping or hind-paw licking as the nociceptive endpoint while 30 seconds was taken as the cutoff point (to avoid any tissue damage).

2.7. Statistical and histological analyses

All data are reported as mean changes in body weight and were compared across treatments and time points. Statistical analysis of the differences between treatment groups was assessed with one-way or two-way analysis of variance followed by Bonferroni test using Prism software (Graph-Pad, San Diego, CA). Significance was set at $P < 0.05$.

At the conclusion of the experiments, each rat was injected with 0.5 μ l of cresyl violet, anesthetized and perfused transcardially with 0.9% isotonic saline, followed by phosphate-buffered saline (PBS) 4% paraformaldehyde (pH 7.4). The brain was removed, stored in the same fixative for 4 h, kept in 20% sucrose overnight, and cut into 20- μ m sections on a freezing microtome. Each coronal section was mounted according to standard histological procedures (Benamar et al., 2004) and the site of injection was verified by locating the dye (Fig 2 C).

3. RESULTS

3.1 Physical dependence to opioid methadone and buprenorphine

Rats were injected with buprenorphine at 9:00 a.m. daily for 7 and 14 consecutive days at doses ranging from 0.5–3 mg/kg (Fig. 1A and 1C). In agreement with Previous study (Cowan, 1995), only animals injected daily with 3 mg/kg of buprenorphine for 14 days showed any sign of withdrawal, which was weight loss that became apparent on day 5 of withdrawal (Fig 1C, $P < 0.001$; two-way ANOVA with repeated measures (body weight $\times$ time), followed by Bonferroni post-test). For comparison purposes we examine physical dependence to methadone (another opioid medication used to manage physical dependence to opioid). The cessation of methadone (5 mg/kg s.c.) after 7 days of daily injection caused a significant body weight loss (Fig. 1B, $P < 0.001$). Other withdrawal signs such as (e.g., wet dog shakes) were observed after the discontinuation of methadone (data not shown). Finally, we examined the effect of buprenorphine at a dose of 5 mg/kg for 7 days. As shown in Fig 1A, the cessation of this opioid after 7 days of daily injection did not induce any withdrawal drop in body weight, which indicates that the earlier development of physical dependence to methadone is not due to the fact that we used a higher dose of methadone compared to buprenorphine.

3.2 The effect of gp120 IIIB on the physical dependence to buprenorphine

Previously it has been shown that intrathecal or epineural injection of gp120 induce pain response (Milligan et al., 2001; Herzberg and Sagen, 2001). First we examined whether gp120 IIIB (133–250 ng) by itself has any effect on pain threshold using the hot-plate test. Gp120 at doses of 133 ng had no effect on pain threshold (Fig 2A, $P > 0.05$) confirming previous data (Chen et al., 2011). Following its injection at a dose of 150 ng, the hot-plate latency was slightly decreased (8.8 ± 0.6 s) compared to control group (10.11 ± 0.4 s). At doses of 250 ng, gp120 produced a significant reduction of hot-plate latency (6.62 ± 0.7 s compared to control (10.11 ± 0.4 s) (Fig 2A, $F_{3,8} = 7.58$, $P < 0.001$). Therefore, to allow a clear data analysis and interpretation, this viral protein at a dose of 133 ng was used to examine its effect on the development of physical dependence. Gp120 was administered into the PAG on days 1, 7 and 14 during the development of physical dependence to

buprenorphine. Gp120 did not precipitate or enhance the withdrawal weight loss (Fig. 2 B). At the end of the experiment, the rats were subjected to histological verification of cannula placements (Benamar et al., 2010). Only data from animals in which the site of injection was clearly located within the ventral and ventro-lateral PAG were included in the studies (Fig 2 C).

Because the physical dependence to methadone occurred earlier than that to buprenorphine even in the absence of gp120 action (Figs 1B and 1C), and a single injection of gp120 into PAG diminished the analgesic effect of methadone (Benamar et al., 2011), we did not examine the effect of gp120 on the physical dependence to methadone.

4. DISCUSSION

The effects of HIV-1 infection on neuronal systems are associated with the entry of the virus into the brain, which occurs soon after the initial infection (Nathanson et al., 1994). While evidence of neuronal infection by HIV-1 is lacking, it is well known that components of the virus such as the coat protein gp120 can bind to, and signal via, neuronal chemokine receptors. Within the brain, gp120, the surface envelope protein used by the virus to gain access into immune cells, has been implicated as a neurotoxic factor in HIV-infected individuals. Gp120 has been detected in the brains of HIV-1-infected individuals, localized by immunohistochemistry to microglia and multinucleated giant cells (Jones et al., 2000). Neurodegeneration and gliosis similar to that seen in patients with HIV-associated dementia (HAD) was found in CNS-targeted gp120 transgenic mice (Mucke et al., 1995; Toggas et al., 1994). To examine whether the presence of gp120 in the brain interferes with the physical dependence to buprenorphine, gp120 has been infused directly into PAG, one of the brain areas involved in physical dependence to opioid, and spontaneous withdrawal behaviors monitored following the discontinuation of the chronic administration of this opioid. Gp120 did not enhance or precipitate the withdrawal-induced weight loss associated with the discontinuation of buprenorphine.

Compared to methadone or morphine, buprenorphine is a partial mu-opioid agonist, which shows high affinity for, and slow dissociation from, mu-opioid receptors. This maintenance of homeostasis may help to counter the development of an overt withdrawal syndrome, and may explain the lower risk of development of physical dependence to buprenorphine compared to methadone. Recently, we found that the infusion of either stromal cell-derived growth factor-1alpha (SDF-1 α /CXCL12) or gp120 into the brain diminished the acute function of morphine and methadone but did not affect buprenorphine (Benamar et al., 2010, 2011; Chen et al., 2011; Palma et al., 2011). The present results show that such a finding extends to the chronic effect of buprenorphine, and indicates that gp120 or the neuro-inflammatory response associated with the presence of this viral protein in the PAG did not promote the development of physical dependence to buprenorphine.

In summary, the present data demonstrate 1) that the physical dependence to methadone occurs earlier compared to buprenorphine, and 2) the presence of gp120 in the brain, a condition associated with HIV-1 infection, did not enhance or precipitate the withdrawal-induced weight loss associated with the discontinuation of buprenorphine. These data

suggest buprenorphine as the better therapeutic option to manage opioid dependence in opioid-using/abusing HIV patients.

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- The physical dependence to methadone occurs earlier compared to buprenorphine
- The presence of gp120 in the brain, a condition associated with HIV-1 infection, did not enhance or precipitate the withdrawal-induced weight loss associated with the discontinuation of buprenorphine
- These data suggest buprenorphine as the better therapeutic option to manage opioid dependence in opioid-using/abusing HIV patients.

Fig. 1.

Fig. 1A. Body weight changes during buprenorphine treatment (day 1–7) and discontinuation (day 8–14). Rats received either saline or buprenorphine (0.5–3 mg/kg, s.c.). Each point represents the mean $\pm$ SEM. N, number of animals.

Fig. 1B. Body weight changes during methadone treatment (day 1–7) and discontinuation (day 8–14). Rats received either saline or buprenorphine (3–5 mg/kg, s.c.). Each point represents the mean $\pm$ SEM. *** $P < 0.001$

Fig. 1C. Body weight changes during buprenorphine treatment (day 1–14) and discontinuation (day 14–28). Rats received either saline or buprenorphine (3 mg/kg, s.c.). Each point represents the mean $\pm$ SEM. *** $P < 0.001$

Fig. 2.

Fig. 2A. The effect of gp120IIIIB on pain threshold. Gp120 (133–250 ng, PAG) or an equivalent volume of vehicle was microinjected at time zero. N, number of rats. Each point represents the mean $\pm$ SEM. Mean response before injection was as follows: $\blacksquare = 10.11 \pm 0.10$ second $\blacktriangle$ (s); $\bullet = 9.87 \pm 0.20$ s; $\bullet = 10.1 \pm 0.11$ s and $\blacktriangledown = 10.09 \pm 0.08$ s. *** $P < 0.001$.

Fig. 2B. Effect of gp120IIIIB (133 ng, PAG) on physical dependence buprenorphine. Each point represents the mean $\pm$ SEM.

Fig 2C. Histological illustration of the distribution of some individual sites of microinjection in the PAG.