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*Original Article*

# Effects of $GABA_B$ receptor blockade on lateral habenula glutamatergic neuron activity following morphine injection in the rat: an electrophysiological study

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## *Abstract*

**Background and purpose:** The lateral habenula (LHb), a key area in the regulation of the reward system, exerts a major influence on midbrain neurons. It has been shown that the gamma-aminobutyric acid (GABA)-ergic system plays the main role in morphine dependency. The role of GABA type B receptors ($GABA_BRs$) in the regulation of LHb neural activity in response to morphine, remains unknown. In this study, the effect of $GABA_BRs$ blockade in response to morphine was assessed on the neuronal activity in the LHb.
**Experimental approach:** The baseline firing rate was recorded for 15 min, then morphine (5 mg/kg; s.c) and phaclofen (0, 0.5, 1, and 2 µg/rat), a $GABA_BRs$' antagonist, were microinjected into the LHb. Their effects on firing LHb neurons were investigated using an extracellular single-unit recording in male rats.
**Findings/Results:** The results revealed that morphine decreased neuronal activity, and $GABA_BRs$ blockade alone did not have any effect on the neuronal activity of the LHb. A low dose of the antagonist had no significant effect on neuronal firing rate, while blockade with doses of 1 and 2 µg/rat of the antagonist could significantly prevent the inhibitory effects of morphine on the LHb neuronal activity.
**Conclusion and implications:** This result indicated that $GABA_BRs$ have a potential modulator effect, in response to morphine in the LHb.

***Keywords:*** Extracellular single-unit recording; $GABA_B$ receptors; Lateral habenula; Morphine.

## INTRODUCTION

Long-term use of morphine as a chronic pain reliever causes dependence and tolerance (1). Opioids can affect the feeling of reward and pleasure through the mesolimbic dopamine system, which includes the ventral tegmental area (VTA), substantia nigra, ventral striatum, prefrontal cortex, and the nucleus accumbens (2). The cellular mechanism of morphine is through the mu-opioid receptor, and several pieces of evidence have shown an association between mu-opioid receptor activation in the lateral habenula (LHb) and the potentiation of morphine effects (3,4).

The LHb includes glutamatergic neurons (5) and several gamma-aminobutyric acid (GABA) inhibitory interneurons (6), having an important function in aversive states, reward processing, and addiction (7). The GABAergic system has an essential role in the central nervous system, and it has been involved also in morphine dependence (8,9) and the rewarding effects of opioids in the VTA (10-12). GABAergic neurons mediate their inhibitory effects through three chief GABA receptors (GABARs) subtypes: termed metabotropic GABA type B receptors ($GABA_BRs$) and $GABA_A$/$GABA_C$ receptors that belong to the ionotropic receptor family of receptors. The $GABA_A/_C$ receptors are ligand-gated Cl-channel, and the $GABA_BRs$ are associated with the $K^+$ channel through the G protein (13,14), responsible for the fast and slow inhibitory response when activated by GABA, respectively (15).

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Electrophysiological studies have demonstrated that drugs of abuse such as morphine and cocaine inhibit LHb neurons (4,16). It has already been identified that GABAergic receptors induce tonic inhibition in the firing of single neurons (17). Also, GABARs' antagonists affected both the firing pattern and spontaneous activity firing rate of neurons in several brain nuclei (18). The other studies also showed the application of ethanol and GABARs' antagonist accelerated the firing rate of LHb neurons (19)

It has already been shown that there is a probable role for GABARs within the LHb (20), but the function and mechanisms in the reward circuit, in particular in response to morphine, in terms of electrophysiology remains unclear. We decided to elucidate the effect of GABA$_B$R blockade on the neuronal activity in LHb, following the systemic application of morphine, using an extracellular recording, because the number of GABA$_B$R in this nucleus is high and their physiological role is unknown, also the effect of blockade of these receptors on LHb neuronal firing rate has not been investigated. On the other hand, there are few reports about the effects of morphine on the neuronal firing rate of this nucleus (4).

## MATERIALS AND METHODS

### *Subjects*

Our subjects were male Wistar rats (250-300 g, prepared from Isfahan University of Medical Sciences, Isfahan, Iran). The animals were kept under controlled temperature and 12/12-h light-dark cycle conditions, with free access to water and food.

We designed our protocols according to the Animal Ethics Committee of Isfahan University of Medical Sciences under Ethic No. IR.mui.MED.REC.1397.244 and the care and use of animals for experimental procedures and use of laboratory animals (National Institutes of Health Publication No. 85-23), revised 2010.

### *Drugs*

Morphine (21,22), urethane, and phaclofen, as a selective GABA$_B$R antagonist (23), were daily and freshly dissolved in 0.9% saline for injection (Table 1).

### *Surgery and electrophysiology*

Rats were deeply anesthetized with urethane (1.6 g/kg, i.p) (24) and after exposing the skull, through stereotactic surgery a hole was drilled for positioning of a double-barrel micropipette (one for drug microinjection and another for recording), into the LHb (AP = -3.7 mm; L= ±0.8 mm; DV = -5.3 mm) (25). The body temperature of the animals was maintained at 37 °C. The recording electrodes were sharp glass micropipettes (1-3 µm) filled with 2 M sodium chloride solution (26). Using an analog to the digital data acquisition and the related software, (eLab; Science Beam Institute, Iran), signals were filtered (300 Hz to 3 kHz bandpass), digitized, analyzed, and presented as a rate histogram. Neurons with a firing rate of < 20 spikes/s and a spike duration > 3 ms were selected. According to electrophysiological characteristics (27-29), we presumed that our target neurons were glutamatergic (Fig. 1A and B).

**Table 1.** Drugs, drug doses, and animal groups used in the present study (n = 6-7).

| Drugs | Doses |
|---|---|
| Morphine (Darou Pakhsh, Iran) | 5 mg /kg |
| Phaclofen (Sigma-Aldrich, Germany) | 0.5, 1, 2 µg/rat |
| Urethane (Sigma-Aldrich, Germany) | 1.6 g/kg |

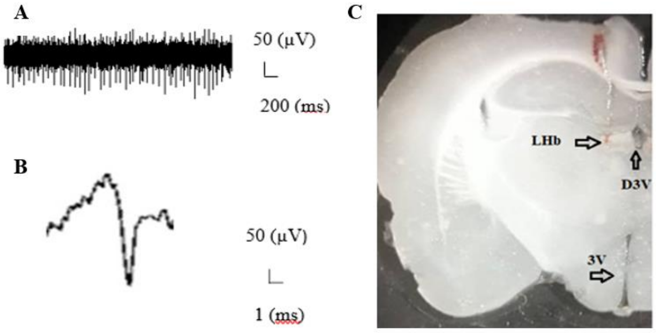

**Fig. 1.** (A) A representative pattern of neuronal electrical activity recorded from the LHb; (B) an expanded waveform of a spike recorded from an LHb neuron; and (C) coronal photomicrograph of the recording and microinjection site in the LHb. 3V, 3rd ventricle; D3V, dorsal 3rd ventricle; LHb, lateral habenula.

After ensuring a steady state, recording started and after 15 min, morphine was injected (5 mg/kg; s.c.). Then, 45 min later different doses of phaclofen (0.5, 1, and 2 µg/0.3 µL) were microinjected, and the recording continued for another 60 min (Fig. 2A). In the control groups, saline was microinjected as a vehicle. In each group, 12 to 18 neurons were evaluated in 6 to 7 rats.

### *Intra-LHb infusions*

To inject the drug into the LHb, the micropipette for drug microinjection was connected to a 1.0-µL glass Hamilton syringe with a short polyethylene tube.

### *Histological verification*

At the end of the study for histological verification of the place of electrodes, rats were perfused transcardially with formalin (10%), and the brains were kept in formalin for 2 days and then sectioned coronally (50 µm thickness; Fig. 1C) (25).

### *Data analysis*

The results were analyzed using SPSS software (version 23). The alterations of mean firing rates were analyzed by repeated measure analysis of variance (ANOVA), the percentage of changes by the one-way ANOVA, and a Tukey test and unpaired Student's *t*-test. All data were expressed as mean ± SEM (n = 6-7 rats). Differences with $P < 0.05$ were considered significant.

## RESULTS

### *LHb neuronal response to morphine*

After ensuring the stability of neuronal activity and baseline recording (15 min), morphine was injected subcutaneously and 45 min later, saline or antagonist was microinjected into the LHb. Morphine (5 mg/kg) had inhibitory effects on the majority of LHb neurons concerning the baseline activity, compared to the saline group (unpaired t-test, -65.34 ± 4.7; 3.26 ± 1.77 respectively; Fig. 2B).

### *LHb neuronal response to intra-LHb injection of saline or phaclofen*

Subcutaneous injection of saline did not induce significant changes in the neuronal activity in LHb, also saline or phaclofen (1 and 2 µg/0.3 µL) microinjection into the LHb did not change the firing rate (spike/s) of the neurons, compared to pre-injection (Fig. 3A-C).

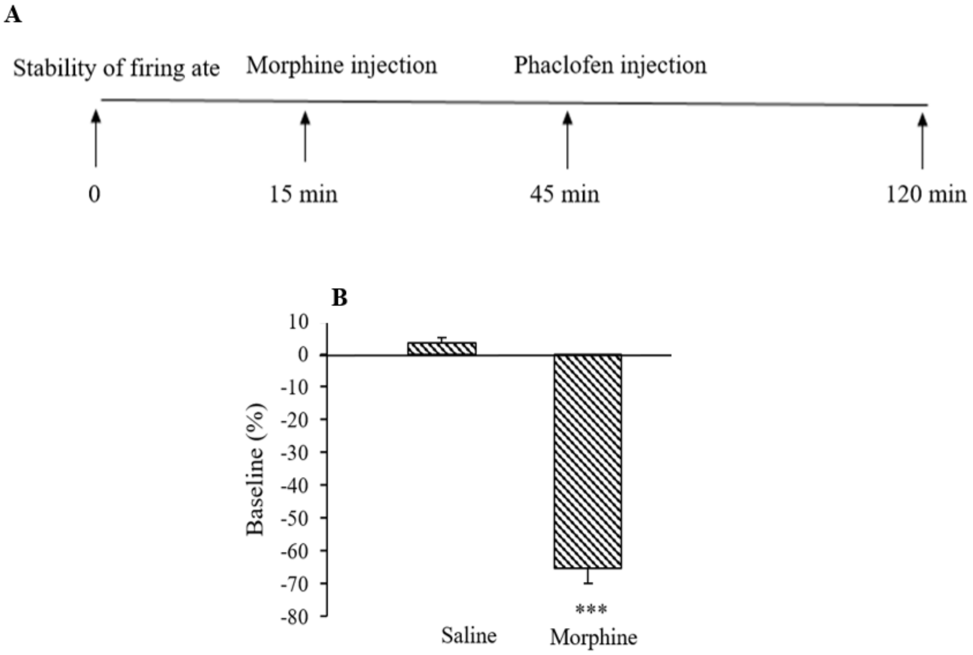

**Fig. 2.** Effect of morphine systemic administration, on the lateral habenula neuronal activity and study protocol. (A) Experimental timeline; **(**B) morphine (5 mg/kg; s.c.) or saline was injected 15 min after a steady state and the recording continued for another 105 min to evaluate the effect of morphine on the lateral habenula neuronal activity with respect to the baseline (unpaired t-test, n = 114 neurons). ***$P < 0.001$ Indicate the significant difference.

In all morphine-treated groups, there was a significant decrease in neuronal firing (spike/s) after morphine injection (*P* < 0.001, Fig. 3D), and intra-LHb injection of saline did not affect this decreasing trend and a significant difference was observed, compared to the baseline (*P* < 0.001, Fig. 3B). Microinjection of phaclofen with doses of 0.5 and 1 µg/0.3 µL into the LHb did not prevent this decreased firing rate (spike/s) (Fig. 3E and F), but 2 µg/0.3 µL of phaclofen increased the neuronal activity of LHb and brought it back to the baseline (Fig. 3G; repeated-measure ANOVA).

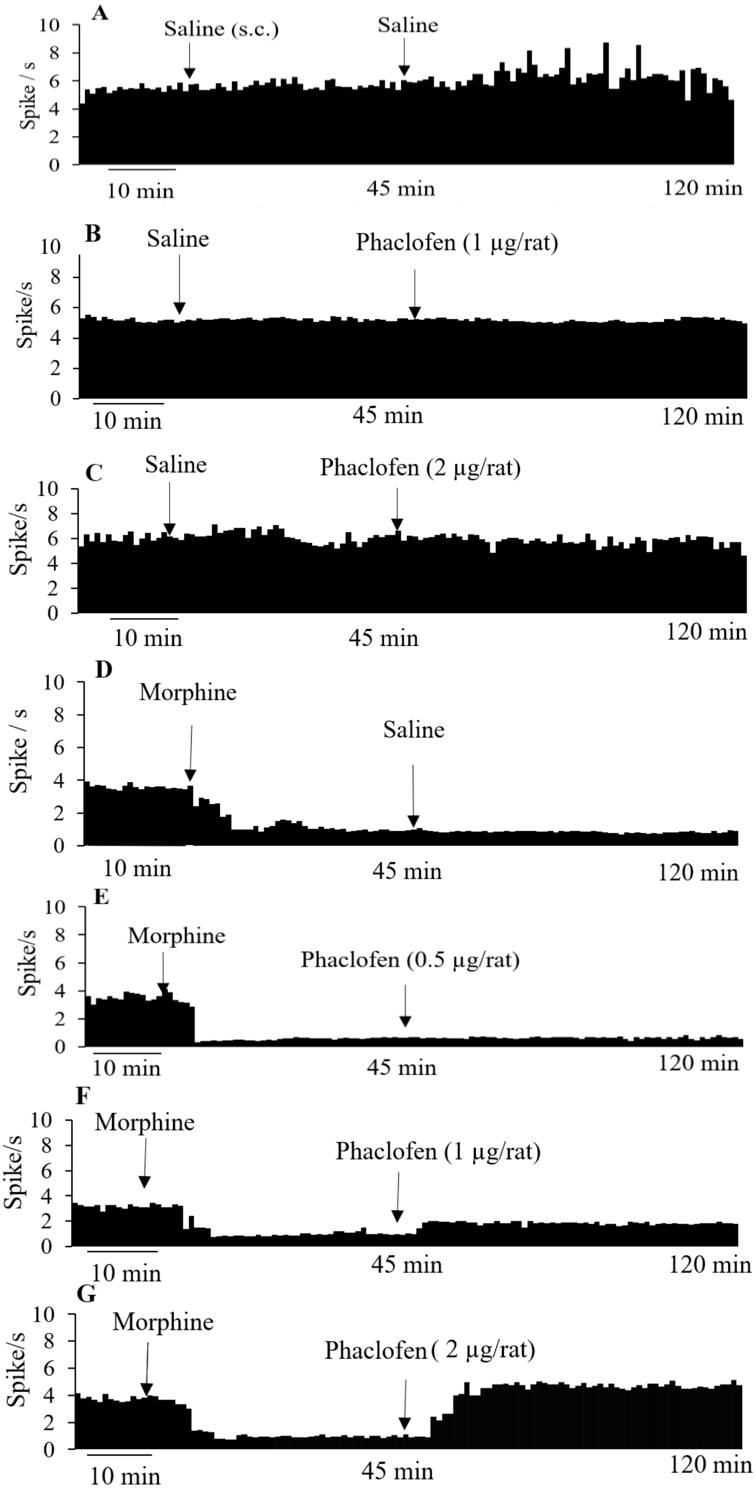

**Fig. 3.** Histograms represent the spike frequency of the entire recording (120 min) of all neurons. Effects of GABA$_B$R antagonist, on the lateral habenula neuronal activity after morphine (5 mg/kg) systemic administration. (A) Saline-saline; (B and C) saline-GABA$_B$R antagonist (phaclofen: 1 and 2 µg/0.3 µL, respectively); (D) morphine-saline; (E-G) morphine-GABA$_B$R antagonist (phaclofen: 0.5, 1, and 2 µg/0.3µL, respectively). GABA$_B$R, gamma-aminobutyric acid receptors receptor.

### *Mean neuronal responses of LHb to block GABABR following administration of morphine*

Intra-LHb injection of saline (10.31 ± 3.19) or phaclofen (1 and 2 µg/0.3 µL and 5.45 ± 4.48, 9.06 ± 4.07; respectively), following subcutaneous injection of saline, did not affect the firing rate (spike/s) of neurons. Morphine administration alone significantly reduced the neuron firing rate (spike/s), compared to the saline group, and after saline microinjection into the LHb, the decrement continued (-85.6 ± 3.49). Also, phaclofen in the morphine-treated rats with a dose of 0.5 µg/0.3 µL, had no significant effect on the reduced firing rate (spike/s) of neurons, induced by morphine injection (-87.78 ± 2.78), but phaclofen in the morphine-treated rats with a dose of 1 µg/0.3 µL, enhanced the neuronal activity concerning the morphine-saline group (-53.71 ± 4.365); however, the firing rate (spike/s) did not return to the level of neuronal activity in the saline group and there was a significant decrease compared to the saline group; while 2 µg/0.3 µL of phaclofen in the morphine-treated rats increased the neuronal activity compared to the morphine and saline groups (40.54 ± 13.63; Fig. 4).

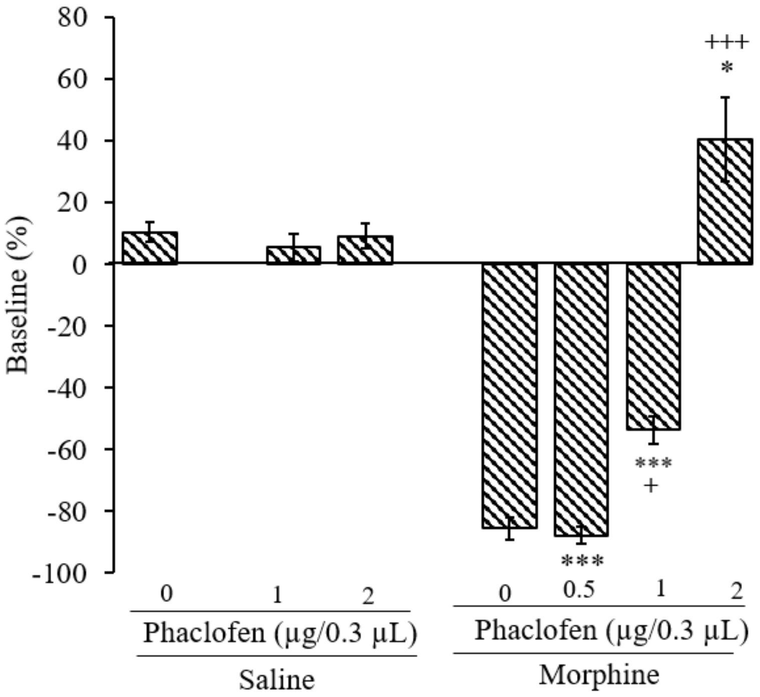

**Fig. 4.** Effects of GABA$_B$R blockade on the lateral habenula neuronal activity. Morphine was injected 15 min after a steady state. Then, 45 min later different doses of phaclofen (0.5, 1, and 2 µg/0.3 µL) were microinjected and the recording continued for another 60 min. Effects of microinjection of GABA$_B$R on the lateral habenula neuronal activity with respect to the baseline morphine-treated rats. Data are expressed as mean ± SEM; n = 6-7 .*$P < 0.05$ and ***$P < 0.001$ indicate significant differences compared to the control group; $^+P < 0.05$, $^{+++}P < 0.001$ versus the morphine group. GABA$_B$R, gamma-aminobutyric acid receptors receptor.

## DISCUSSION

LHb obtains GABAergic afferents from the VTA (20) and basal ganglia (30), but there is little evidence for the presence of GABA-ergic neurons in the LHb (31-33). The large majority of LHb neurons are glutamatergic (32,34) and they receive strong glutamatergic afferents from numerous brain areas, including the anterior cingulate, entopeduncular nucleus (4), and medial prefrontal cortex (20). Therefore, in the present study, according to the spike duration and firing rate, the selected neurons were assumed to be glutamatergic (27,29,32,35). It has been suggested that glutamatergic neurons in LHb and GABAergic neurons in VTA play a major role in VTA functional activity and thus local inhibition of mesolimbic dopamine neurons, respectively (34). The rewarding effects of morphine significantly depend on GABAergic neurons and their excitatory and inhibitory afferents (5).

Our results showed that morphine decreases the LHb-neuronal activity (Figs. 2 and 3B), which plays an important role in mediating the reward effects of addictive compounds, including morphine (3,4). It has been already shown that LHb neurons respond to systematic injections of morphine with a decreased firing rate (4). Morphine has been postulated to decrease the firing frequency by hyperpolarizing neurons through two distinct synaptic mechanisms (1) postsynaptic hyperpolarization or (2) inhibition of presynaptic glutamate release (4). The results of the present study showed that blocking $GABA_BRs$ prevents morphine-induced decrement of neuronal activity in LHb, especially at the high dose of the antagonist (Fig. 3Fand G and Fig. 4). Probably due to the number of $GABA_BRs$ and their distribution in this area despite high doses, the low dose of the antagonist had no significant effect on neuronal firing rate (Fig. 3E). It is interesting that the blockade of $GABA_BRs$ alone in the saline group, did not have any effect on the activity of neurons (Fig. 3A, C, D and, Fig. 4), so they may not be involved in the basal activity on their own, but may mediate morphine action. Also, it has already been identified that GABARs' antagonists affected both the firing pattern and spontaneous activity firing rate of neurons in several brain nuclei and induced an increase in the firing rate (18).

It has been reported that the LHb displays a high expression of $GABA_BRs$, possibly on glutamatergic neurons (36), but the functions of $GABA_BRs$ in this nucleus in both physiological and pathological conditions remain poorly characterized. It has been demonstrated that these receptors in the LHb can control baseline neuronal activity (37), as well as a vast number of neuronal properties, involving excitability and synaptic strength (38). Previous evidence has shown that $GABA_BRs$ may inhibit LHb neurons by inhibiting adenylyl cyclase and mediating post-synaptic hyperpolarization (36,39,40). Dysregulation of the function of these receptors has been implicated in several disorders including anxiety, depression, and addiction, where the role of LHb is crucial (36,38). Also, the potential role of $GABA_BRs$ in the control of functions of LHb neurons, especially in the context of aversion and reward, remains to be investigated.

Studies have demonstrated the effectiveness of the intra-LHb blockade of GABARs on place preference behavior (41,42); these effects are probably due in part to the imbalance between glutamatergic and GABAergic LHb neurons, which can lead to damage to reward circuits and pathological complications following morphine use (31). It has been reported that a shift towards the reduction in GABA neurotransmission in the LHb leads to enhance the excitability of GABAergic neurons in the tail of the ventral tegmental area, and finally results in the loss of the rewarding effects of morphine (4,5,42).

## CONCLUSIONS

Our data showed that the firing rate of neurons of LHb was significantly suppressed following systemic injection of morphine. Although blockade of $GABA_BRs$ in the saline group did not induce any change in the firing rate, microinjection of phaclofen in morphine-receiving groups was able to prevent morphine-induced firing rate reduction. Our findings suggest that $GABA_BRs$ probably play a mediating role in rewarding responses to morphine. However, further studies are needed to identify the signaling pathways and intracellular mechanisms involved in this process.

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### *Conflict of interest statements*

The authors declared no conflict of interest in this study.

### *Authors' contributions*

P. Reisi. Contributed to the concept of the study, design, the definition of intellectual content, statistical analysis, data analysis, manuscript editing, and manuscript review; H. Alaei contributed to the concept of the study, manuscript editing, and the definition of intellectual content. E. Amohashem did the literature search and experimental studies, acquire the data, prepared the manuscript, and statistically analyzed the data. The finalized article was approved by all authors.

## REFERENCES

1. Ueda H, Ueda M. Mechanisms underlying morphine analgesic tolerance and dependence. Front Biosci (Landmark Ed). 2009;14(14):5260-5272. DOI: 10.2741/3596.
2. Tuominen L, Tuulari J, Karlsson H, Hirvonen J, Helin S, Salminen P, *et al*. Aberrant mesolimbic dopamine-opiate interaction in obesity. Neuroimage. 2015;122:80-86. DOI: 10.1016/j.neuroimage.2015.08.001.
3. Kim J, Ham S, Hong H, Moon C, Im HI. Brain reward circuits in morphine addiction. Mol Cells. 2016;39(9):645-653. DOI: 10.14348/molcells.2016.0137.
4. Margolis EB, Fields HL. Mu opioid receptor actions in the lateral habenula. PloS One. 2016;11(7):e0159097,1-11. DOI: 10.1371/journal.pone.0159097.
5. Flanigan M, Aleyasin H, Takahashi A, Golden SA, Russo SJ. An emerging role for the lateral habenula in aggressive behavior. Pharmacol Biochem Behav. 2017;162:79-86. DOI: 10.1016/j.pbb.2017.05.003.
6. Bianco IH, Wilson SW. The habenular nuclei: a conserved asymmetric relay station in the vertebrate brain. Philos Trans R Soc B Biol Sci. 2009;364(1519):1005-1020. DOI: 10.1098/rstb.2008.0213
7. Matsumoto M, Hikosaka O. Representation of negative motivational value in the primate lateral habenula. Nat Neurosci. 2009;12(1):77-84. DOI: 10.1038/nn.2233.
8. Taylor AMW, Castonguay A, Ghogha A, Vayssiere P, Pradhan AA, Xue L, *et al*. Neuroimmune regulation of GABAergic neurons within the ventral tegmental area during withdrawal from chronic morphine. Neuropsychopharmacology. 2016;41(4):949-959. DOI: 10.1038/npp.2015.221.
9. Ghamkharinejad G, Marashi SH, Foolad F, Javan M, Fathollahi Y. Unconditioned and learned morphine tolerance influence hippocampal-dependent short-term memory and the subjacent expression of GABA-A receptor alpha subunits. PloS One. 2021;16(9):e0253902,1-22. DOI: 10.1371/journal.pone.0253902
10. Suzuki T, Nurrochmad A, Ozaki M, Khotib J, Nakamura A, Imai S, et al. Effect of a selective $GABA_B$ receptor agonist baclofen on the μ-opioid receptor agonist-induced antinociceptive, emetic and rewarding effects. Neuropharmacology. 2005;49(8):1121-1131. DOI: 10.1016/j.neuropharm.2005.06.009.
11. Galaj E, Han X, Shen H, Jordan CJ, He Y, Humburg B, *et al*. Dissecting the role of GABA neurons in the VTA *versus* SNr in opioid reward. J Neurosci. 2020;40(46):8853-8869. DOI: 10.1523/JNEUROSCI.0988-20.2020.
12. Bouarab C, Thompson B, Polter AM. VTA GABA neurons at the interface of stress and reward. Front Neural Circuits. 2019;13:78,1-12. DOI: 10.3389/fncir.2019.00078.
13. Enna SJ. The GABA receptors. In: Enna SJ, Möhler H, editors. The GABA receptors: Springer; 2007. pp. 1-21. DOI: 10.1007/978-1-59745-465-0_1.
14. Auteri M, Zizzo MG, Serio R. GABA and GABA receptors in the gastrointestinal tract: from motility to inflammation. Pharmacol Res. 2015;93:11-21. DOI: 10.1016/j.phrs.2014.12.001.
15. Chebib M, Hinton T, Schmid KL, Brinkworth D, Qian H, Matos S, *et al*. Novel, potent, and selective GABAC antagonists inhibit myopia development and facilitate learning and memory. J Pharmacol Exp Ther. 2009;328(2):448-457. DOI: 10.1124/jpet.108.146464.
16. Dougherty PM, Qiao J, Wiggins R, Dafny N. Microiontophoresis of cocaine, desipramine, sulpiride, methysergide, and naloxone in habenula and parafasciculus. Exp Neurol. 1990;108(3):241-246. DOI: 10.1016/0014-4886(90)90129-G.
17. Hutt A. The population firing rate in the presence of GABAergic tonic inhibition in single neurons and application to general anaesthesia. Cogn Neurodyn. 2012;6(3):227-237. DOI: 10.1016/10.1007/s11571-011-9182-9.
18. Paladini C, Tepper J. Neurophysiology of substantia nigra dopamine neurons: modulation by GABA and glutamate. In: Steiner H, Tseng KY, editors. Handbook of behavioral neuroscience. Elsevier; 2016. pp. 335-60. DOI: 10.1016/B978-0-12-802206-1.00017-9.
19. Zuo W, Wang L, Chen L, Krnjević K, Fu R, Feng X, *et al*. Ethanol potentiates both GABAergic and glutamatergic signaling in the lateral habenula. Neuropharmacology. 2017;113(Pt A):178-187.

DOI: 10.1016/j.neuropharm.2016.09.026.

20. Batalla A, Homberg JR, Lipina TV, Sescousse G, Luijten M, Ivanova SA, *et al*. The role of the habenula in the transition from reward to misery in substance use and mood disorders. Neurosci Biobehav Rev. 2017;80:276-285. DOI: 10.1016/j.neubiorev.2017.03.019.
21. Berkowitz BA. The relationship of pharmacokinetics to pharmacological activity: morphine, methadone and naloxone. Clin Pharmacokinet. 1976;1(3):219-230. DOI: 10.2165/00003088-197601030-00004.
22. Lee MR, Yu SC, Hwang BH, Chen CY. Determining morphine in biologic fluids of rats by gas chromatography–mass spectrometry. Anal Chim Acta. 2006;559(1):25-29. DOI: 10.1016/j.aca.2005.11.059.
23. Amohashemi E, Reisi P, Alaei H. Lateral habenula electrical stimulation with different intensities in combination with GABAB receptor antagonist reduces acquisition and expression phases of morphine-induced CPP. Neurosci Lett. 2021;759:135996,1-6. DOI: 10.1016/j.neulet.2021.135996.
24. Dolatabadi LK, Reisi P. Acute effect of cholecystokinin on short-term synaptic plasticity in the rat hippocampus. Res Pharm Sci. 2014;9(5):331-336.
25. Paxinos G, Watson C. The rat brain in stereotaxic coordinates. 6th ed. Elsevier; 2006. pp: 57-67
26. Fartootzadeh R, Alaei H, Reisi P. Mutual assistance of nucleus accumbens cannabinoid receptor-1 and orexin receptor-2 in response to nicotine: a single-unit study. Res Pharm Sci. 2021;16(2):173-181. DOI: 10.4103/1735-5362.310524.
27. Kowski A, Veh R, Weiss T. Dopaminergic activation excites rat lateral habenular neurons *in vivo*. Neuroscience. 2009;161(4):1154-1165. DOI: 10.1016/j.neuroscience.2009.04.026.
28. Sotty F, Danik M, Manseau F, Laplante F, Quirion R, Williams S. Distinct electrophysiological properties of glutamatergic, cholinergic and GABAergic rat septohippocampal neurons: novel implications for hippocampal rhythmicity. J Physiol. 2003;551(3):927-943. DOI: 10.1111/j.1469-7793.2003.00927.x.
29. Weiss T, Veh R. Morphological and electrophysiological characteristics of neurons within identified subnuclei of the lateral habenula in rat brain slices. Neuroscience. 2011;172:74-93. DOI: 10.1016/j.neuroscience.2010.10.047.
30. Golden SA, Heshmati M, Flanigan M, Christoffel DJ, Guise K, Pfau ML, *et al*. Basal forebrain projections to the lateral habenula modulate aggression reward. Nature. 2016;534(7609):688-692. DOI: 10.1038/nature18601.
31. Barker DJ, Miranda-Barrientos J, Zhang S, Root DH, Wang HL, Liu B, *et al*. Lateral preoptic control of the lateral habenula through convergent glutamate and GABA transmission. Cell Rep. 2017;21(7):1757-1769. DOI: 10.1016/j.celrep.2017.10.066.
32. Lecca S, Meye FJ, Mameli M. The lateral habenula in addiction and depression: an anatomical, synaptic and behavioral overview. Eur J Neurosci. 2014;39(7):1170-1178. DOI: 10.1111/ejn.12480.
33. Zhang L, Hernández VS, Swinny JD, Verma AK, Giesecke T, Emery AC, *et al*. A GABAergic cell type in the lateral habenula links hypothalamic homeostatic and midbrain motivation circuits with sex steroid signaling. Transl Psychiatry. 2018;8(1):50,1-14. DOI: 10.1038/s41398-018-0099-5.
34. Stamatakis AM, Stuber GD. Activation of lateral habenula inputs to the ventral midbrain promotes behavioral avoidance. Nat Neurosci. 2012;15(8):1105-1107. DOI: 10.1038/nn.3145.
35. Kim U, Chang SY. Dendritic morphology, local circuitry, and intrinsic electrophysiology of neurons in the rat medial and lateral habenular nuclei of the epithalamus. J Comp Neurol. 2005;483(2):236-250. DOI: 10.1002/cne.20410.
36. Meye FJ, Lecca S, Valentinova K, Mameli M. Synaptic and cellular profile of neurons in the lateral habenula. Front Hum Neurosci. 2013;7:860,1-7. DOI: 10.3389/fnhum.2013.00860.
37. Lecca S, Pelosi A, Tchenio A, Moutkine I, Lujan R, Hervé D, *et al.* Rescue of GABA B and GIRK function in the lateral habenula by protein phosphatase 2A inhibition ameliorates depression-like phenotypes in mice. Nat Med. 2016;22(3):254-261. DOI: 10.1038/nm.4037.
38. Lüscher C, Slesinger PA. Emerging roles for G protein-gated inwardly rectifying potassium (GIRK) channels in health and disease. Nat Rev Neurosci. 2010;11(5):301-315. DOI: 10.1038/nrn2834
39. Geisler S, Andres KH, Veh RW. Morphologic and cytochemical criteria for the identification and delineation of individual subnuclei within the lateral habenular complex of the rat. J Comp Neurol. 2003;458(1):78-97. DOI: 10.1002/cne.10566.
40. Lüscher C, Jan LY, Stoffel M, Malenka RC, Nicoll RA. G protein-coupled inwardly rectifying K+ channels (GIRKs) mediate postsynaptic but not presynaptic transmitter actions in hippocampal neurons. Neuron. 1997;19(3):687-695. DOI: 10.1016/S0896-6273(00)80381-5.
41. Good CH, Wang H, Chen YH, Mejias-Aponte CA, Hoffman AF, Lupica CR. Dopamine D4 receptor excitation of lateral habenula neurons *via* multiple cellular mechanisms. J Neurosci. 2013;33(43):16853-16864. DOI: 10.1523/JNEUROSCI.1844-13.2013.
42. Lu YG, Wang L, Chen JL, Zhu J, Meng XY, You ZD, *et al.* Projections from lateral habenular to tail of ventral tegmental area contribute to inhibitory effect of stress on morphine-induced conditioned place preference. Brain Res. 2019;1717:35-43. DOI: 10.1016/j.brainres.2019.03.026.