ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 14, Issue 7, 2021

Online - 2455-3891
Print - 0974-2441

Research Article

POSSIBLE INFLUENCE OF LOXOPROFEN IN LIPOPOLYSACCHARIDE INDUCED ALTERATIONS IN SUCROSE INTAKE IN CHRONIC MILD STRESS MODEL IN MICE

KUNDU SMITA S, DIGVIJAYSINH G RANA*

Department of Pharmacology, Babaria Institute of Pharmacy, Vadodara, Gujarat, India. Email: dgrana3755@yahoo.com

Received: 28 February 2021, Revised and Accepted: 02 June 2021

ABSTRACT

Objective: The objective of the present study was to evaluate the influence of Loxoprofen in sucrose intake in the absence and presence of Lipopolysaccharide in chronic mild stress model of depression in mice.

Methods: There was a measurement of sucrose intake in chronic mild stress model (CMS), consisting of 21 days stress schedule in which mice were subjected to the treatment of Loxoprofen (16.8 mg/kg, *p.o.*) with or without treatment of lipopolysaccharide (LPS) (0.5 mg/kg *i.p.*) for the past 14 days.

Results: The result of the present study indicated that mice treated with Venlafaxine and Loxoprofen showed a significant increase in the sucrose intake in stressed mice in chronic mild stress model. LPS-treated mice presented a decrease in sucrose intake when compared to controls. Similarly, Venlafaxine and Loxoprofen in the presence of LPS could increase the sucrose intake as compared to LPS treated stressed mice.

Conclusion: The results of the present study showed that Loxoprofen could influence LPS induced alterations in sucrose intake in mice in chronic mild stress model. It can also indicate the possible anti-depressant effect of Loxoprofen in mice subjected to chronic mild stress model of depression, having its possible implication in future treatment of depression.

Keywords: Chronic mild stress, Depression, Lipopolysaccharide, Loxoprofen, Mice.

 DOI: http://dx.doi.org/10.22159/ajpcr.2021v14i7.41249. Journal homepage: https://innovareacademics.in/journals/index.php/ajpcr

INTRODUCTION

Depression is a major mood disorder which is characterized by altered mood with recurrent thoughts of suicide. It has been surveyed that depression may become the second-most disabling disease after cardiovascular disease by 2020 [1]. Various investigations have been reported for the explorations of pathophysiology of depression. Despite such growing evidences, there exist many limitations of current antidepressants treatment [2].

Considering such reports of limitations, it can be proposed that there is a need for more exploration of pathophysiology of depression. Further, the role of inflammation in the neurobiology of depression has received a considerable amount of research attention in the past few years [3-7]. It has been reported that there is an abnormal prostaglandin E levels in depression [8-10]. Various studies reported that that there was an elevated level of prostaglandins (PGs) especially PGE_2 in depression [8-9,11]. Further, neuro-inflammation may be contributed by PGE_2 as concluded from various preclinical studies [12].

Several studies indicated that there was an increase in levels of prostaglandin E_2 (PGE_2) in depressed individuals [8-10,13-14]. Further, a high prostaglandin E_2 (PGE_2) levels have repeatedly been described in major depression [15]. Further, an upregulation of cyclooxegenase-2 (COX-2) is associated with increased PGE_2 levels and neuronal apoptosis [16]. COX-2 inhibitors inhibited the PGE_2 synthesis, suggesting a potential positive role in depression [17-18]. Several studies have reported that antidepressant drugs could inhibit PGE_2 synthesis including MAO inhibitors [19-22] and Tricyclic antidepressants [23].

Loxoprofen is a drug of non-steroidal anti-inflammatory drugs category with anti-inflammatory properties. It has been found that Loxoprofen remarkably decreased the PGE_2 levels in regions of brain in some preclinical studies [24].

Considering above mentioned reports, it can be hypothesized that Loxoprofen may affect depressive behaviors in experimental animals. Hence, it has been proposed to study the effect of Loxoprofen on sucrose intake in chronic mild stress model of depression in mice with or without presence of Lipopolysaccharide.

METHODS

Animals

Swiss albino male mice were obtained from Zydus Research Centre, Moraiya, Ahmedabad. They were housed under standard condition with free access to food and water, under 12:12 h light: dark cycle. Mice were allowed to acclimatize for 07 days before the initiation of behavioral tests. Each animal was used only once (n=6 animals per group). The experiments were performed after the protocol for experimental design was approved with protocol no. BIP/IAEC/2018/06 by the Institutional Animal Ethics Committee (IAEC) of Babaria Institute of Pharmacy. The experiments were conducted according to the guidelines of the Committee for the Purpose of Control and Supervision of Experiment on Animals (CPCSEA)

Drugs

Venlafaxine [VEN] as triple monoamine reuptake inhibitor was administered in the dose of 4 mg/kg i.p. [25]. Lipopolysaccharide [LPS] was administered in the dose of 0.5mg/kg *i.p.* [26]. Loxoprofen [LOX] was dissolved in 0.5% CMC solution [24]. Venlafaxine was dissolved in normal saline solution (0.9% NaCl) [25]. LPS was dissolved in phosphate-buffered saline (PBS) solution [26].

Chronic mild stress (CMS)

Training of animals for sucrose intake test

At first, mice were trained to consume 2% sucrose solution for 1 week. After a period of 1 week, baseline test was carried out for sucrose intake (two tests per week) over a time period of 18 days for all mice. These tests consist of a 3 h of food and water deprivation period followed by

giving sucrose solution for 1 h. Intake was then measured by weighing the bottles before and after each test. After the baseline value of the sucrose intake, all mice were further divided into subgroups according to the matched baseline value of sucrose intake [27].

Groups of animals based on matched baseline value of sucrose intake

Total sixty mice were subjected to sucrose intake test at an interval of day 0, day 3, day 6, day 9, day 12, day 15, and day 21. In this set of experiment, 18 mice were housed under normal condition. They were subdivided into three groups: Group I received saline and served as vehicle control- I (VC-I), Group II received 0.5% CMC served as vehicle control- II (VC-II), and Group III received Lipopolysaccharide (0.5mg/kg *i.p.*) (LPS).

Rest of 42 mice were housed under chronic mild stress conditions and were divided as follows: Group IV received stress and saline (10 ml/kg *p.o.*) and served as STR-I, Group V received stress and 0.5% CMC (10ml/kg *p.o.*) and served as STR-II, Group VI received stress and Venlafaxine (4 mg/kg *i.p.*), Group VII received stress and Loxoprofen (16.8 mg/kg *p.o.*), Group VIII received stress and Lipopolysaccharide (0.5mg/kg *i.p.*), Group IX received stress and Lipopolysaccharide (0.5mg/kg *i.p.*) and Venlafaxine (4 mg/kg *i.p.*), and Group X received stress and Lipopolysaccharide (0.5mg/kg *i.p.*) and Loxoprofen (16.8 mg/kg *p.o.*)

Stress schedule followed in CMS

In this set of experiment, the stress schedule for producing chronic mild stress was followed according to the already reported stressor schedule [27]. The stress scheme included various stressors which included 3 periods of water and food deprivation of 5 h immediately before the sucrose tests, two periods of intermittent illumination, two periods (7 and 12 h) of 45° cage tilting, one additional 16 h period of water deprivation, one 12 h period in a soiled cage (adding 100 ml water in the bedding), and three periods (7, 9, and 12 h) of low intensity stroboscopic illumination (150 flashes/min). These stressors were scheduled every day for a total period of 21 days.

Treatment schedule of drugs administration in CMS

Sucrose intake was initially measured at day 0, day 3, and day 6. After day 7, mice were then subjected to the treatment of drugs as per group schedule for 14 days as mentioned above. Sucrose intake was later be measured consequently at day 9, day 12, day 15, day 18, and day 21.

Statistical analysis

All quantified data were expressed as mean±S.E.M. for the indicated analyses. Statistical comparisons were performed by one-way ANOVA followed by the Tukey's Test. $p<0.05$ was considered as significant. All statistical analyses were performed using approved statistical software (Sigmastat software, Systat Software Inc, San Jose, CA, USA.).

RESULTS

Effect of stress schedule on sucrose intake in stressed mice

There was a decrease in sucrose intake in mice of STR-I and STR-II group when compared with mice of VC-I and VC-II group, respectively. There was a significant decrease in sucrose intake in LPS treated stressed mice as compared to LPS treated normal mice (Table 1).

Effect of Venlafaxine and Loxoprofen on sucrose intake in stressed mice without treatment of LPS

The treatment of Venlafaxine showed significant increase in sucrose intake when compared to STR-I. The treatment of Loxoprofen showed significant increase in sucrose intake when compared to STR-II. The treatment of Loxoprofen showed a non-significant increase in sucrose intake when compared to Venlafaxine (Table 1).

Effect of Venlafaxine and Loxoprofen on sucrose intake in LPS treated stressed mice

The treatment of Venlafaxine showed significant increase in sucrose intake when compared to LPS treated stressed mice on day 21. Similarly, the treatment of Loxoprofen also significantly increases the sucrose intake when compared to LPS treated stressed mice on day 21 (Table 1).

DISCUSSION

The results of the present investigations indicated the potential antidepressant-like effect of Loxoprofen in chronic stress model of depression in mice. However, such results did not answer the question of whether behavioral sampling information of any of animal models of depression could reliably be compared with clinical outcome of depression since screening for the antidepressant agents through the animal models of depression always demand the accurate validation of models with its greater construct validity, predictive validity, and face validity. Chronic mild stress was reported as one of the chronic models of depression with higher construct validity which can establish empirical relationship between the feature being modeled and depression in humans [28]. However, despite higher constructive validity of CMS model which might enable us to correlate the clinical symptoms, more evidences are required to be furnished to confirm whether Loxoprofen may affect the behavior in animals.

The mechanism by which Loxoprofen indicated a significant anti-depressant like action in chronic mild stress model remains to be elucidated. However, the previously reported inhibitory action of Loxoprofen on PGE_2 synthesis may show potential role of PGE_2 in mediating the anti-depressant effect of Loxoprofen in animal models of depression. It can also be possible that antidepressant effect of Loxoprofen at specified dose as mentioned in the present study may be achieved by the inhibition of PGE_2 in brain. It is also possible that such Loxoprofen induced PGE_2 inhibition may be responsible for the inhibition of synthesis of inflammatory mediators.

Table 1: Effect of Venlafaxine and Loxoprofen on sucrose intake in stressed mice

Group No	Treatment	Sucrose intake (g/kg)							
		Day 00	Day 03	Day 06	Day 09	Day 12	Day 15	Day 18	Day 21
1	VC-I	154.6 ± 19.80	159.61±19	105.09±9.07	72.96±9	92.75±9.9	89.6 ±11.7	98.9±5.7	146.7±19.2
2	VC-II	140.58±8.84	166.69±22.87	72.79±10	81.82±7.74	93.28±8.48	52.88±6.65	51.30 ±7.25	56.58±11.58
3	LPS	175.72±15.40	173.25±14.16	68.55±8.82	106.4±21.37	61.6±4.68	67.5±9.62	90±9.16	82.66±14.64
4	STR-I	149.48±17.19	160.92±36.68	66.01±12.2*	83.53±11.61	58±11.2*	54.4±10.5*	56.9±7.87*	51.10±8.32*
5	STR-II	139.81± 34.98	154.52±27.64	74.77±11.22	60.56±5.35^{+}	53.74±9.11^{+}	23.46±2.7^{+}	22.67±7.13^{+}	20.27±5.27^{+}
6	STR+VEN	96.095±11.99	128.98±19.55	68.20±11.02	73.33±11.75	67.5±11	85.6±8.34$^{\$}$	84.7±4.85$^{\$}$	86.67±10.1$^{\$}$
7	STR+LOX	187.63±24.15	151.57±21.05	43.45±7.14	82.81±8.06$^{@}$	84.69±7.07$^{@}$	82.75±11.9$^{@}$	70.82±14.5$^{@}$	74.78±14.8$^{@}$
8	STR+LPS	132.81±21.77	203.05±11.38	97.51±15.52	76.25±7.57	64.1±5.62	45.4±10.9	40.6±8.87	31.5±9.99$^{\#}$
19	STR+LPS+VEN	125.26±7.12	114.52±22.73	60.98±15.17	63.16 ±13.77	44.7±7.57	60.3±5.82	34.6±8.74	61.25±8.4$^{£}$
10	STR+LPS+LOX	109.54±14.81	205.71±16.33	65.36±15.57	57.30±12.92	66.32±13.74	59.73 ± 10.57	35.31±7	60.67±8$^{£}$

Each column expressed as Mean±SEM of six animals after respective treatments. Data were analysed by One-way Analysis variance (ANNOVA) followed by Tukey's test. *$p<0.05$ when compared with VC-I, $^{+}p<0.05$ when compared with VC-II, $^{\#}p<0.05$ when compared with LPS, $^{\$}p<0.05$ when compared with STR-I, $^{@}p<0.05$ when compared with STR-II, $^{£}p<0.05$ when compared with STR+LPS

Although we were unable to measure brain PGE_2 levels in brain, further research work is suggested to examine if Loxoprofen induced alteration in brain PGE_2 levels may affect the molecular mechanism of depression such as alterations in expression of brain derived neurotrophic factor (BDNF) gene.

Although, the behavior sampling data of chronic mild stress model show the potential anti-depressant action of Loxoprofen in mice, further work is required for more exploration of the present investigations.

Regardless of the previous studies, these are the first results for the potential anti-depressant like effect of Loxoprofen in chronic mild stress model of depression in mice, having its potential implication in the pathophysiology and treatment of depression in future.

CONCLUSION

The results of the present study indicated that Loxoprofen could influence LPS induced alterations in sucrose intake in mice in chronic mild stress model. It also indicated the possible anti-depressant like effect of Loxoprofen in mice subjected to chronic mild stress model of depression, having its possible implication in future treatment of depression.

ACKNOWLEDGMENTS

The authors are grateful management team of Babaria Institute of Pharmacy, Vadodara, India, for providing necessary facilities for this work. Also thankful to Zydus Research Centre, Ahmedabad, for providing animals and all other pharmaceutical companies for providing gratis sample of drug for this research.

AUTHOR'S CONTRIBUTIONS

Dr. Digvijay Rana designed the research study; Ms. Smita S. Kundu performed the study; Dr. Digvijay Rana prepared the manuscript; all authors approved the final submitted version of the manuscript.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHOR'S FUNDING

This research received no external funding.

REFERENCES

1. Simon GE. Social and economic burden of mood disorders. Biol Psychiatry 2003;54:208-15.
2. Zajecka JM. Clinical issues in long-term treatment with antidepressants. J Clin Psychiatry 2000;61:20-5.
3. Patel A. The role of inflammation in depression. Psychiatr Danub 2013;25 Suppl 2:216-23.
4. Farooq RK, Asghar K, Kanwal S, Zulqernain A. Role of inflammatory cytokines in depression: Focus on interleukin-1β. Biomed Rep 2017;6:15-20.
5. Felger JC, Lotrich FE. Inflammatory cytokines in depression: Neurobiological mechanisms and therapeutic implication. J Neurosci 2013;29:199-229.
6. Jangpangi D. Depression and inflammation: Pathophysiology and therapeutic implications. CHRISMED J Health Res 2016;3:155-60.
7. Noto C, Rizzo LB, Mansur RB, McIntyre RS, Maes M, Brietzke E. Targeting the inflammatory pathway as a therapeutic tool for major depression. Neuroimmunomodulation 2014;21:131-9.
8. Lieb J, Karmali R, Horrobin DF. Elevated levels of PGE_2, and the thromboxane B_2 in depression. Prostaglandins Leukot Med 1983;10:361-7.
9. Calabrese JR, Skwerer RG, Barna B, Gulledge AD, Valenzuela R, Butkus A, *et al*. Depression, immunocompetence, and prostaglandins of the E series. Psychiatry Res 1986;17:41-7.
10. Linnoila M, Whorton AR, Rubinow DR, Cowdry RW, Ninan PT, Waters RM. CSF prostaglandin levels in depressed and schizophrenic patients. Arch Gen Psychiatry 1983;40:405-6.
11. Fritz M, Klawonn AM, Nilsson A, Singh AK, Lazarus M, Löfberg A, *et al*. Prostaglandin dependent modulation of dopaminergic neurotransmission elicits inflammation-induced aversion in mice. J Clin Invest 2016;126:695-705.
12. Brenneis C, Coste O, Altenrath K, Angioni C, Schmidt H, Schuh CD, *et al*. Anti-inflammatory role of microsomal prostaglandin E synthase-1 in a model of neuroinflammation. J Biol Chem 2010;286:2331-42.
13. Ohishi K, Ueno R, Nishino S, Sakai T, Hayaishi O. Increased level of salivary prostaglandins in patients with major depression. Biol Psychiatry 1988;23:326-34.
14. Nishino S, Ueno R, Ohishi K, Sakai T, Hayaishi O. Salivary prostaglandin concentrations: Possible state indicators for major depression. Am J Psychiatry 1989;146:365-8.
15. De Paiva VN, Lima SN, Fernandes MM, Soncini R, Andrade CA, Giusti-Paiva A. Prostaglandins mediate depressive-like behaviour induced by endotoxin in mice. Behav Brain Res 2010;215:146-51.
16. Li RC, Row BW, Gozal E, Kheirandish L, Fan Q, Brittian KR, *et al*. Cyclooxygenase 2 and intermittent hypoxia-induced spatial deficits in the rat. Am J Respir Crit Care Med 2003;168:469-75.
17. Müller N, Schwarz MJ, Dehning S, Douhe A, Cerovecki A, Goldstein-Müller B, *et al*. The cyclo-oxygenase 2 inhibitor celecoxib has therapeutic effects in major depression: Results of a double blind, randomized, placebo controlled, add on pilot study to reboxetine. Mol Psychiatry 2006;11:680-4.
18. Teeling JL, Cunningham C, Newman TA, Perry VH. The effect of non-steroidal anti-inflammatory agents on behavioural changes and cytokine production following systemic inflammation: Implications for a role of COX-1. Brain Behav Immun 2010;24:409-19.
19. Lee RE. The influence of psychotropic drugs on prostaglandin biosynthesis. Prostaglandins 1974;5:63-8.
20. Bekemeier H, Giessler AJ, Vogel E. Influence of MAO inhibitors, neuroleptics, morphine, mescaline, divascan, aconitine, and pyrogenes on prostaglandin biosynthesis. Pharmacol Res Comm 1977;9:587-98.
21. Fjalland B. Influence of various substances on prostaglandin biosynthesis by guinea pig chopped lung. J Pharm Pharmacol 1976;28:683-9.
22. Hong SL, Carty T, Deykin D. Tranylcypromine and 15-hydroperoxy arachidonate affect arachidonic acid release in addition to inhibition of prostaglandin synthesis in calf aortic endothelial cells. J Biol Chem 1980;255:9538-40.
23. Mtabaji JP, Manku MS, Horrobin DF. Actions of the tricyclic anti depressant clomipramine on responses to pressor agents. Interactions with prostaglandin E_2. Prostaglandins 1977;14:273-81.
24. Futaki N, Harada M, Sugimoto M, Hashimoto Y, Honma Y, Arai I, *et al*. The importance of brain PGE_2 inhibition versus paw PGE2 inhibition as a mechanism for the separation of analgesic and antipyretic effects of Lornoxicam in rats with paw inflammation. J Pharm Pharmacol 2009;61:607-14.
25. Thomas J, Khanam R, Vohora D. Augmentation of effect of venlafaxine by folic acid in behavioral paradigms of depression in mice: Evidence of serotonergic and pro-inflammatory cytokine pathways. Pharmacol Rep 2016;68:396-403.
26. Mello BS, Monte AS, McIntyre RS, Soczynska JK, Custódio CS, Cordeiro RC, *et al*. Effects of doxycycline on depressive-like behavior in mice after lipopolysaccharide (LPS) administration. J Psychiatr Res 2013;47:1521-9.
27. Chen Y, Wang HD, Xia X, Kung HF, Pan Y, Kong LD. Behavioural and biochemical studies of total furocoumarins from seeds of *Psoralea corylifolia* in the chronic mild stress model of depression in mice. Phytomedicine 2007;14:523-9.
28. Abramsom LY, Seligmen ME. Modeling psychopathology in the laboratory: History and rationale. In: Maser JP, Seligman ME, editors, Psychopathology; Experimental Models. San Francisco: Freeman Inc.; 1978. p. 1-26.