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ANTIDEPRESSANT-LIKE ACTIVITY OF LUTEIN-ZEAXANTHIN IN MICE: EVIDENCE FOR INVOLVEMENT OF MONOAMINERGIC AND OPIOID SYSTEMS

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Abstract

Aims: The antinociceptive-antidepressant effects of lutein-zeaxanthin (LZ) have been studied in rats. However, there is no data on the antidepressant-like activity of LZ in mice or humans. Hence the current study was undertaken to evaluate acute and chronic antidepressant-like action of lutein-zeaxanthin in mouse forced swim test (FST) and tail suspension test (TST), and to investigate possible mechanism of action. **Methods:** Adult female albino mice 2–3 months old were used in the study. LZ was administered in four doses – 2.5, 5, 10 and 20 mg/kg, and antidepressant-like activity was tested using both FST and TST, with fluoxetine as positive control. Effect on locomotor activity and of chronic administration of drug (dosing for 14 days) was also investigated. Mechanism of action was determined using different receptor antagonists – p-chlorophenylalanine (pCPA), pindolol, ketanserin, ondansetron, prazosin, yohimbine and naloxone. **Results:** Antidepressant activity was seen with LZ 5, 10 and 20 mg/kg in both FST and TST ($p<0.001$), which was comparable to fluoxetine. There was no effect on the locomotor activity. Reversal of antidepressant-like action of LZ was seen with pCPA (serotonin synthesis inhibitor), pindolol (5-$HT_{1A/1B}$ receptor antagonist), ketanserin ($5HT_{2A/2C}$ receptor antagonist), prazosin (α_1-adrenoceptor antagonist), yohimbine (α_2-adrenoceptor antagonist) and naloxone (nonselective opioid receptor antagonist), but not with ondansetron ($5HT_3$ receptor antagonist). **Conclusion:** Lutein-zeaxanthin produces an antidepressant-like action in the preclinical level, by modulating serotonergic (5-$HT_{1A/1B}$ and 5-$HT_{2A/2C}$ receptors), noradrenergic (α_1 and α_2 receptors) and opioid pathways.

Key Words: Antidepressant, Forced swim test, Lutein, Serotonin, Tail suspension test, Zeaxanthin.

Introduction

Major depression is a common psychiatric disorder, widely distributed in the population and causing significant disability to the patient. [1] According to the World Health Organization report, by the year 2020, depression will be the second leading cause of disability.

[2] In this era, a multitude of antidepressant drugs are available in the market for the treatment of depression. Though these drugs produce some improvement in the clinical condition of patient, they have a slow onset of action and cause numerous adverse effects due to their broad mechanisms of action. [3] Thus the search for novel drugs for the treatment of major depression is the need of the hour. Most of the current antidepressant drugs act by modulating serotonergic and catecholamine systems. They either directly affect serotonin (5-HT) turnover in the brain, or prevent 5-HT or norepinephrine reuptake and enhance monoaminergic neurotransmission.

[4] Besides these well-known mechanisms of action, there is significant data indicating the activation of the opioid system as a mechanism of antidepressant action. [5, 6] Hence it is essential to investigate the probable role of monoaminergic and opioid systems in the antidepressant-like effect of experimental drugs.

Lutein and zeaxanthin are major carotenoid pigments present in human retina. They are xanthophylls which coexist and are synthesized in plants and some micro-organisms; giving corn, saffron, mango, and many other plants and microbes their characteristic colour.

[7] They have antioxidant properties which have been found to afford protection in age-related macular degeneration of eye. [8] It was also reported that zeaxanthin, by enhancing anti-oxidative capacity, prevented cell death both *in vivo* and *in vitro*. [9] Lutein and zeaxanthin have also been found to have a beneficial role in treatment of cardiovascular diseases due to anti-atherogenic effect, and they lower the risk of mortality in Alzheimer's disease. [10, 11]

Major depression has been linked to decreased levels of various endogenous antioxidant compounds, and this presents plausible targets for development of newer drugs based on antioxidant compounds. [12] Antinociceptive-antidepressant effects of lutein-zeaxanthin have been studied in rats. [13] However, there is no data on the antidepressant-like activity of lutein-zeaxanthin (LZ) in mice or humans or its possible mechanism of action. Considering this, the current study was undertaken with following objectives: 1.

To evaluate acute and chronic antidepressant-like action of lutein-zeaxanthin in mouse forced swim test (FST) and tail suspension test (TST). 2. To study the possible role of serotonergic, noradrenergic and opioid systems in its antidepressant-like action.

Materials and Methods

Experimental animals:

Adult female albino mice 2–3 months old (weighing 25–30 g) were maintained at 22–25°C, under a 12:12 h light/dark cycle. They were housed six per cage with free access to food and water. All experiments were performed on separate groups of animals (N=6 animals per group) between 9 am and 4 pm, and each animal was used only once in each test. The study protocol was approved by the Institutional Animal Ethics Committee (1081/a/07/CPCSEA/N-10/2014) and all the procedures were performed according to guidelines set by The Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), India. All efforts were made to lessen the suffering and to decrease the number of animals used in the experiments.

Drugs

Drugs used in this study were procured from standard commercial traders. The test drug lutein-zeaxanthin was obtained from Source Naturals Inc., (Santa Cruz, CA, USA). Each 10 mg capsule (5% marigold flower extract) contains 6 mg lutein and 4 mg zeaxanthin. Fluoxetine, p-chlorophenylalanine (pCPA), pindolol, ketanserin, ondansetron, prazosin, yohimbine and naloxone were all of analytical grade and were acquired from Sigma Chemical Co., (St. Louis, MO, USA). Receptor antagonists were dissolved in saline or 1% Tween 80 and administered to mice intraperitoneally, in a constant volume of 10 mg/ml body weight. Appropriate vehicle treated groups were also assessed simultaneously.

Experimental procedure

Evaluation of antidepressant-like effect after acute administration of lutein-zeaxanthin

Animals were administered a single oral dose of saline, lutein-zeaxanthin or fluoxetine and then tested by FST or TST after 60 minutes. LZ was made as a suspension in saline and given at four different doses of 2.5, 5, 10 and 20 mg/kg. The dose selection of lutein-zeaxanthin followed a logarithmic fashion, based on previous studies and a pilot study done in our laboratory. [13] Fluoxetine (20 mg/kg p.o.), a selective serotonin reuptake inhibitor, was administered 60 minutes preceding the tests and was the positive control. [14]

Evaluation of antidepressant-like effect after chronic administration of lutein-zeaxanthin

For the chronic study, mice were given saline, fluoxetine or lutein-zeaxanthin (2.5, 5, 10, 20 mg/kg), orally for 14 days. 60 minutes after the last dose on the 14th day, mice were assessed using FST and TST to check for antidepressant effect.

Evaluation of lutein-zeaxanthin's possible mechanism of antidepressant-like action using FST

Pretreatment of mice was done with different receptor antagonists or their respective vehicles, 30 minutes following which, lutein-zeaxanthin was administered (10 mg/kg, p.o.). Mice underwent FST 60 minutes after lutein-zeaxanthin treatment. Dose and pretreatment period of all antagonists were based on earlier reports. [15-17]

Role of serotonergic system in antidepressant-like effect of lutein-zeaxanthin in FST

Intraperitoneal injection of pCPA (100 mg/kg) was given to mice, once daily for four successive days as pretreatment. 30 minutes after the last injection of pCPA, mice received either a vehicle or lutein-zeaxanthin and underwent FST after another 60 minutes.

[17] To investigate possible involvement of serotonergic receptors in the antidepressant-like effect of lutein-zeaxanthin, animals were pretreated with intraperitoneal injections of pindolol (10 mg/kg, a 5-$HT_{1A/1B}$ receptor antagonist), ketanserin (5 mg/kg, a $5HT_{2A/2C}$ receptor antagonist) or ondansetron (1 mg/kg, $5HT_3$ receptor antagonist). [15-17] After 30 minutes, they received lutein-zeaxanthin or vehicle and underwent FST 60 minutes later.

Role of noradrenergic system in antidepressant-like action of lutein-zeaxanthin in FST

Animals were pretreated with intraperitoneal injections of prazosin (1 mg/kg, an α_1-adrenoceptor antagonist) or yohimbine (1 mg/kg, an α_2-adrenoceptor antagonist). [15] After 30 minutes, they were administered lutein-zeaxanthin or vehicle and were tested by FST after another 60 minutes.

Role of opioid receptors in antidepressant action of lutein-zeaxanthin in FST

Mice were pretreated with intraperitoneal injection of naloxone (1 mg/kg, a nonselective opioid receptor antagonist). [16] 30 minutes following this, they received lutein-zeaxanthin or vehicle and underwent FST after an additional 60 minutes.

Behavioral analysis

Forced swim test (FST)

Test was conducted using the method described by Porsolt with some modifications. [18] A five liter glass cylinder was filled with water to a height of 15 cm, and each mouse was placed in it, and observed for a duration of 6 minutes. Duration of immobility during last 4 minutes of the procedure was only recorded.

Animal was considered immobile when it floated motionless or made only those movements necessary to keep its head above the water surface. Mice were removed and dried with a towel after the procedure. Water was replaced after testing of each animal.

Tail suspension test (TST)

Method described by Steru was used for this test. [19] Acoustically and visually isolated mice were suspended by the tail on a thin horizontal steel rod, 50 cm above the surface with the help of an adhesive tape placed approximately 1 cm from tip of the tail. Immobility time during last 4 minutes of a 6 minute procedure was only recorded. Animals were considered immobile only when they hung passively and were completely motionless.

Locomotor activity of mice using actophotometer

Procedure used for this test was that described by Boissier and Simon with slight modification. [20] All animals were evaluated in this test to exclude any change brought about by the test drug in locomotor activity. Doses of receptor antagonists were selected based on those reported by previous studies. [16, 17, 21] Actophotometer contains a square arena (30×30 cm) with walls that are fitted with photocells just above floor level. The photocells were checked before beginning the experiment. Mice were then individually placed in the arena. After a two minute acclimatization period, frequency of crossing the light beam was recorded automatically for next 6 minutes. [20, 22]

Statistical analysis

Data was recorded in the GraphPad Prism 7.01 software and analyzed using one-way analysis of variance (ANOVA) followed by Tukey's *post hoc* test. A two-way ANOVA followed by Bonferroni's *post hoc* test was done to analyze differences between groups used for investigating mechanism of action. Results have been represented as mean±SD. Statistical significance was set at a probability value less than 0.05 ($p<0.05$).

Results

Effect of acute treatment with lutein-zeaxanthin on immobility time in FST of mice

Effect of acute administration of lutein-zeaxanthin on immobility time of mice in FST is depicted in Fig. 1. From the p*ost hoc* analysis, it was found that doses of 5, 10 and 20 mg/kg of lutein-zeaxanthin decreased immobility time significantly [$F(5,30) = 116.09$, $p<0.001$]. Fluoxetine which served as positive control also significantly decreased ($p<0.001$) immobility time.

Effect of chronic treatment with lutein-zeaxanthin on immobility time in FST of mice

Fig. 2 shows effect of chronic administration of lutein-zeaxanthin on immobility time of mice in FST. Immobility time was significantly reduced by lutein-zeaxanthin at doses of 5, 10 and 20 mg/kg as compared to control group in a dose-dependent fashion [$F(5,30) = 206.84$, $p<0.001$]. It was seen that fluoxetine also caused a significant decrease in immobility time ($p<0.001$).

Effect of acute treatment with lutein-zeaxanthin on immobility time in TST of mice

Evaluation of the effect of acute treatment with lutein-zeaxanthin was done also by TST in mice and doses of 5, 10 and 20 mg/ kg were seen to produce a statistically significant decrease in immobility time [$F(5,30) = 134.67$, $p<0.001$] (Fig. 3). This decrease in immobility time was comparable with that produced by fluoxetine ($p<0.001$).

Effect of chronic treatment with lutein-zeaxanthin on immobility time in TST of mice

It was seen that the chronic administration of lutein-zeaxanthin also significantly decreased the immobility time in TST at doses 5, 10 and 20 mg/kg [$F(5,30) = 106.42$, $p<0.001$] (Fig. 4). Fluoxetine also produced a similar, significant decrease in immobility period ($p<0.001$).

Role of serotonergic system in antidepressant action of lutein-zeaxanthin in FST

Results in Fig. 5A show that pCPA alone did not modify immobility time, while pretreatment of mice with pCPA significantly reversed the action of lutein-zeaxanthin (10 mg/kg, p.o.) on immobility time in FST. Two-way ANOVA showed a significant effect of lutein-zeaxanthin treatment [$F(1,20)=356.4$, $p<0.001$], pCPA pretreatment [$F(1,20)=730.5$, $p<0.001$] and lutein-zeaxanthin×pCPA interaction [$F(1,20)=813.9$, $p<0.001$]. Furthermore, decrease in immobility time produced by lutein-zeaxanthin was blocked by pretreatment with ketanserin (Fig. 5C) [lutein-zeaxanthin treatment: $F(1,20)=222.5$, $p<0.001$, ketanserin pretreatment: $F(1,20)=750.4$, $p<0.001$, lutein-zeaxanthin×ketanserin interaction: $F(1,20)=1048$, $p<0.001$]. The antidepressant-like effect of lutein-zeaxanthin (10 mg/kg, p.o.) was also prevented by pre-treatment of mice with pindolol (Fig. 5D) [lutein-zeaxanthin treatment: $F(1,20)=88.11$, $p<0.001$, pindolol pretreatment: $F(1,20)=435.4$, $p<0.001$, lutein-zeaxanthin×pindolol interaction: $F(1,20)=621.1$, $p<0.001$]; but not with ondansetron (Fig. 5B) [lutein-zeaxanthin treatment: $F(1,20)=1170$, $p<0.001$, ondansetron pretreatment: $F(1,20)=7.4$, $p=0.01$, lutein-zeaxanthin×ondansetron interaction: $F(1,20)=0.38$, $p=0.54$].

Role of noradrenergic system in antidepressant-like action of lutein-zeaxanthin in FST

The reduction in immobility time caused by lutein-zeaxanthin (10 mg/kg, p.o.) in FST was significantly blocked by pretreatment of mice with prazosin (Fig. 6A) or yohimbine (Fig. 6B). Two-way ANOVA showed significant differences in effect of lutein-zeaxanthin [$F(1,20)=183.2$, $p<0.001$], prazosin [$F(1,20)=688.7$, $p<0.001$], and lutein-zeaxanthin×prazosin interaction [$F(1,20)=746.3$, $p<0.001$]. Similarly, it was seen that yohimbine pretreatment blocked the anti-immobility effect of lutein-zeaxanthin [lutein-zeaxanthin treatment: $F(1,20)=122$, $p<0.001$, yohimbine pretreatment: $F(1, 20)=434.1$, $p<0.001$, lutein-zeaxanthin×yohimbine interaction: $F(1,20)=383.5$, $p<0.001$].

Role of opioid receptors in antidepressant-like action of lutein-zeaxanthin in FST

Fig. 7 depicts the effect of pretreatment of mice with naloxone on immobility time in FST. It was observed that naloxone significantly reduced anti-immobility effect caused by lutein-zeaxanthin (10 mg/kg, p.o.). A two-way ANOVA revealed significant differences in effect of lutein-zeaxanthin [$F(1,20)=79.94$, $p<0.001$], naloxone [$F(1,20)=529.5$, $p<0.001$], and lutein-zeaxanthin×naloxone interaction [$F(1,20)=2276$, $p<0.001$].

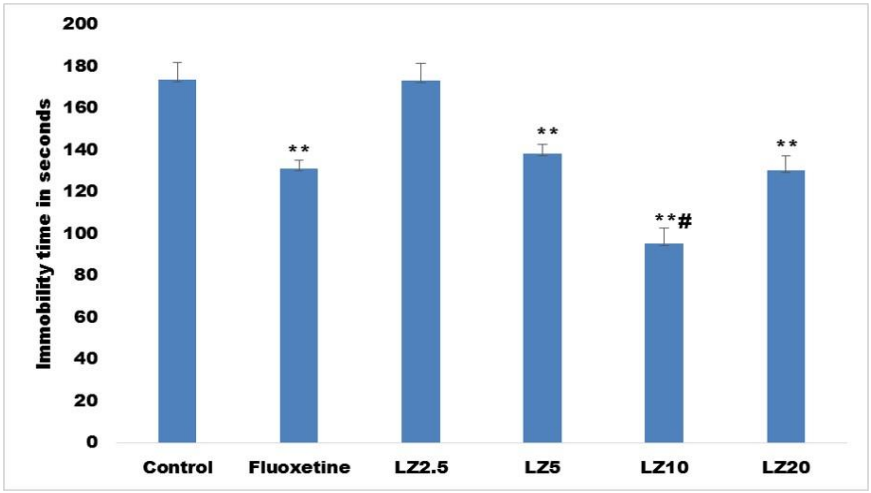
Effect caused by lutein-zeaxanthin on locomotor activity of mice in actophotometer

Results depicted in Table 1 show that neither acute [$F(5,30)=2.107$, $P=0.092$] nor chronic [$F(5,30)= 4.441$, $P=0.391$] administration of lutein-zeaxanthin at any of the four doses (2.5, 5, 10 and 20 mg/kg, p.o.) caused a change in locomotor activity of mice. Similarly, fluoxetine too had no effect on locomotor activity tested using actophotometer.

Table 1: Effect of acute and chronic administration of lutein-zeaxanthin on the locomotor activity in the actophotometer performance of mice.

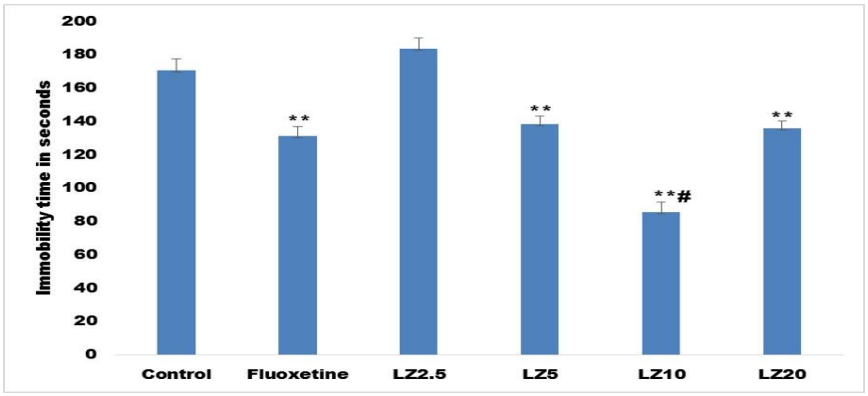
Drug	Number of crossings (mean±SEM)	
	Acute administration	Chronic administration
Control	324.33 ± 11.16	354.5 ± 4.18
Fluoxetine (20 mg/kg)	349.83 ± 16.27	336.67 ± 7.51
Lutein-zeaxanthin (2.5 mg/kg)	300.5 ± 3.43	350.83 ± 7.09
Lutein-zeaxanthin (5 mg/kg)	342.00 ± 10.13	330 ± 4.28
Lutein-zeaxanthin (10 mg/kg)	331.67 ± 10.37	359.17 ± 3.77
Lutein-zeaxanthin (20 mg/kg)	331.50 ± 14.54	350.5 ± 3.58

Fig. 1: Effect of acute administration of lutein-zeaxanthin in forced swim test in mice.



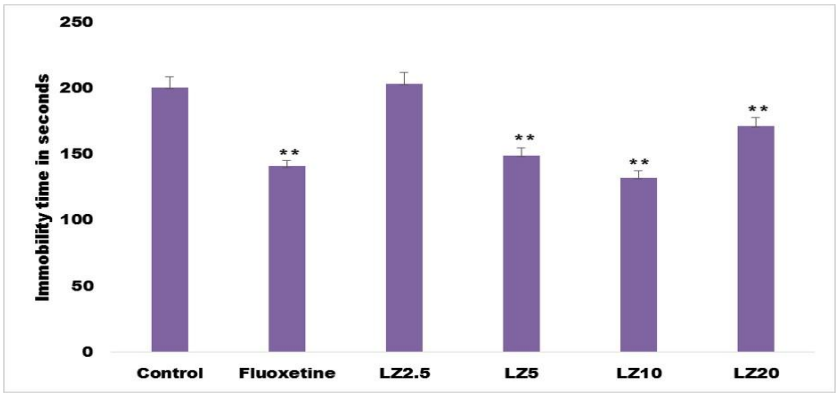
Each column represents mean±S.D. from 6 animals per group. **$p<0.001$ as compared to control group, #$p<0.001$ as compared to fluoxetine group, LZ – lutein-zeaxanthin.

Fig. 2: Effect of chronic administration of lutein-zeaxanthin in forced swim test in mice.



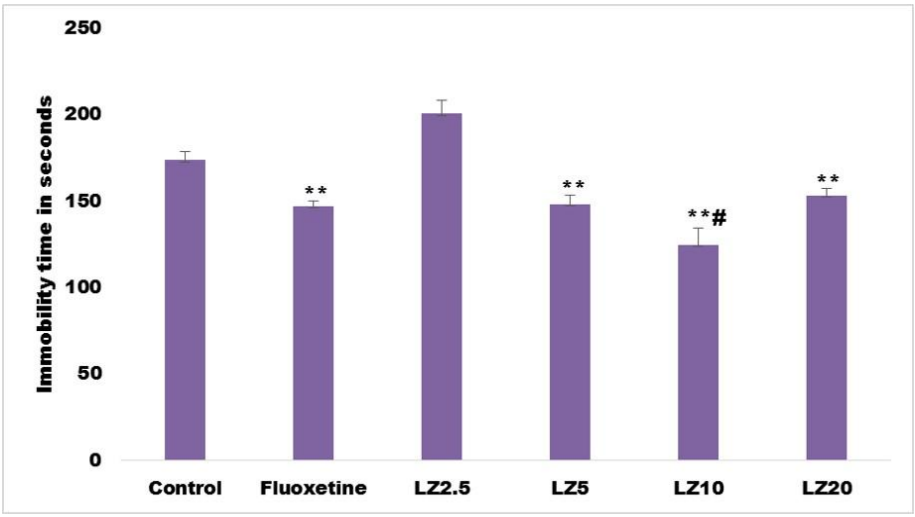
Each column represents mean±S.D. from 6 animals per group. **p<0.001 as compared to control group, #p<0.001 as compared to fluoxetine group, LZ – lutein-zeaxanthin.

Fig. 3: Effect of acute administration of lutein-zeaxanthin in tail suspension test in mice.



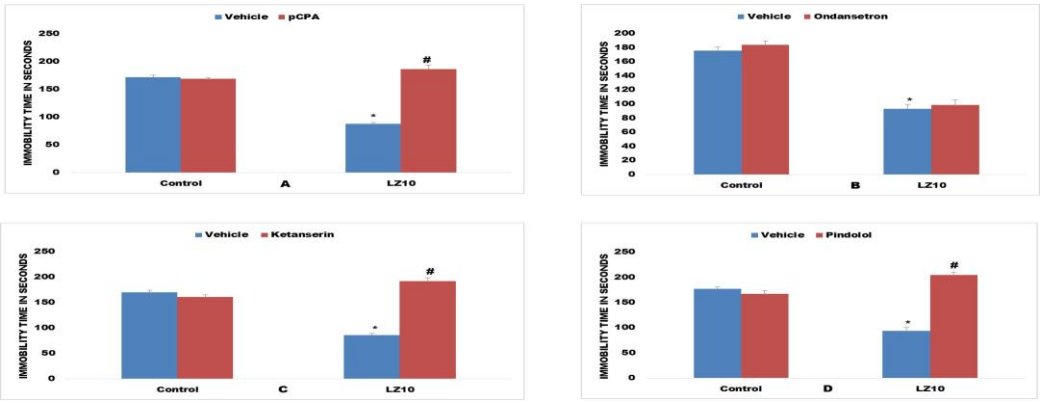
Each column represents mean±S.D. from 6 animals per group. **p<0.001 as compared to control group, LZ – lutein-zeaxanthin

Fig. 4: Effect of chronic administration of lutein-zeaxanthin in forced swim test in mice.



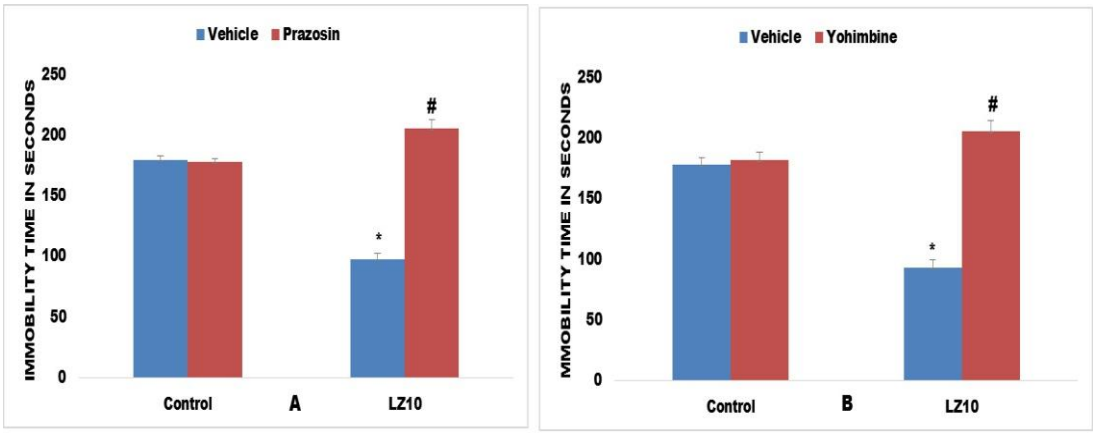
Each column represents mean±S.D. from 6 animals per group. **p<0.001 as compared to control group, #p<0.001 as compared to fluoxetine group, LZ – lutein-zeaxanthin

Fig. 5: Effect of pretreatment with serotonergic antagonists in forced swim test in mice.



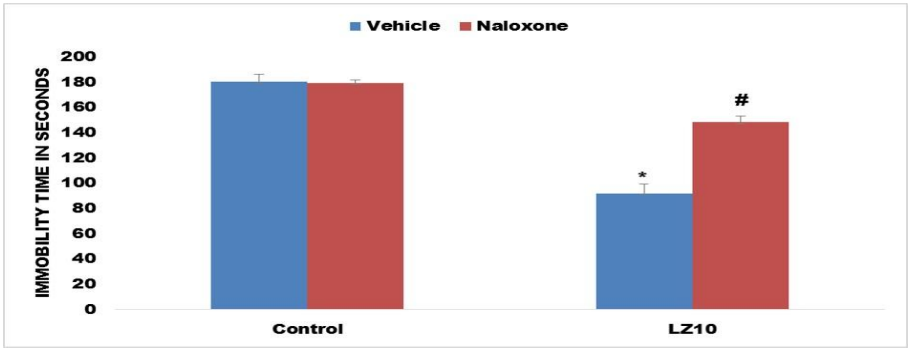
Effect of pretreatment of mice with pCPA (100 mg/kg i.p., panel A), ondansetron (1 mg/kg i.p., panel B), ketanserin (5 mg/kg i.p., panel C) and pindolol (10 mg/kg i.p., panel D) on the immobility time of lutein-zeaxanthin (10 mg/kg p.o.) in the forced swimming test. Each column represents mean±S.D. from 6 animals per group. *p<0.001 as compared to control group, #p<0.001 as compared to LZ alone, LZ – lutein-zeaxanthin

Fig. 6: Effect of pretreatment with adrenoceptor blockers in forced swim test in mice.



Effect of pretreatment of mice with prazosin (1 mg/kg i.p., panel A) and yohimbine (1 mg/kg i.p., panel B) on the immobility time of lutein-zeaxanthin (10 mg/kg p.o.) in the forced swimming test. Each column represents mean±S.D. from 6 animals per group. *p<0.001 as compared to control group, #p<0.001 as compared to LZ alone, LZ – lutein-zeaxanthin

Fig. 7: Effect of pretreatment with naloxone in forced swim test in mice.



Effect of pretreatment of mice with naloxone (1 mg/kg i.p.) on the immobility time of lutein-zeaxanthin (10 mg/kg p.o.) in the forced swimming test. Each column represents mean±S.D. from 6 animals per group. *p<0.001 as compared to control group, #p<0.001 as compared to LZ alone, LZ – lutein-zeaxanthin.

Discussion

The current study aimed at evaluating antidepressant-like activity of lutein-zeaxanthin given by oral route in mice using two predictive models of depression in animals, namely FST and TST. Possible mechanisms by which the drug acted was also explored. The study demonstrated dose-dependent antidepressant-like effect of LZ in both models of depression. It was further observed that antidepressant-like action of LZ was modulated by multiple mechanisms – serotonergic, noradrenergic and opioid pathways. Antinociceptive-antidepressant effects of xanthophylls (lutein, zeaxanthin and lutein-zeaxanthin) were established in Sprague-Dawley rats and they were found to modulate brain monoamines and spinal reactive oxygen species along with modulation of protein and mRNA expression in spinal cord. [13] Results of the current study further support these reports. To the best of our knowledge, this is the first study of its kind to establish antidepressant-like action of lutein-zeaxanthin in mice and to attempt to study the mechanism of action. Among frequently used animal models for screening of antidepressant activity, FST and TST are the foremost. These models are known to be sensitive to these drugs after acute administration. [23, 24] Both tests are similar in their objectives and are based on the fact that when animals are placed in a restricted, inescapable situation, they ultimately cease to struggle and concede to the experimental conditions. [18, 19] The antidepressant-like activity is demonstrated in these models by a reduction in the immobility time of the animals. [18, 25] Moreover, they are important models to study neurobiological mechanisms involved in antidepressant responses and are simple and reliable. [19, 26] These models have been used for screening of antidepressant-like activity of various compounds and plant extracts. [5, 6, 17, 27, 28]. In this study, lutein-zeaxanthin produced an antidepressant-like action on both acute and chronic administration by oral route in both models – FST and TST. The effect thus produced was similar to that produced by the control drug, fluoxetine. Antidepressant-like action was seen with all doses tested in a dose-dependent fashion, except for the lowest – 2.5 mg/kg. Another interesting observation was the U-shaped trend demonstrated by LZ in both test models. This trend has previously been reported by various other studies in endogenous modulatory compounds. [21, 22, 23, 27, 29] However, the reason for this U-shaped trend is obscure and further investigations are necessary for elucidating it. Psychostimulant drugs have been known to increase locomotor activity, thereby reducing immobility time in FST and TST. [27, 29] To rule out such false positive results, locomotor

activity was tested. Anti-immobility effect produced by lutein-zeaxanthin was apparently not related to any motor effects, as animals that were administered the test drug did not reveal excessive ambulation when tested in the actophotometer. This signifies that a psychostimulant effect is not the cause for the decrease in immobility elicited by lutein-zeaxanthin in both models (FST and TST), indicating that antidepressant-like effect of lutein-zeaxanthin is specific. Although a variety of factors are thought to contribute towards development of major depressive disorders, the role of the monoaminergic system in pathophysiology and treatment of depression is well-documented. [30, 31] The monoamine deficiency hypothesis, as suggested by its name, states that the pathology of depression involves a deficit in levels of endogenous monoamine neurotransmitters, mainly serotonin and noradrenaline. [32] Similar studies in the past have reported involvement of monoaminergic system in antidepressant-like action of compounds. [23, 28, 33] In accordance with above-mentioned findings, an attempt was made in the current study to investigate action of lutein-zeaxanthin on monoaminergic system using FST model. In order to explore the mediation of LZ activity by the serotonergic system, various antagonists were used, namely pCPA, pindolol, ketanserin and ondansetron. Previous studies have shown that pCPA given at the dose of 100mg/kg for four consecutive days resulted in depletion of endogenous serotonin without any effect on noradrenergic or dopaminergic levels. [17] In the current study, it was seen that pretreatment with pCPA blocked the effect of LZ on immobility time in FST, implicating a role of serotonergic system in antidepressant-like effect of LZ. Pretreatment with pindolol and ketanserin reversed anti-immobility effect of LZ, thereby indicating that action of this drug is brought about, atleast in part, by an interaction with 5-$HT_{1A/1B}$ and 5-$HT_{2A/2C}$ receptors. However, administration of ondansetron failed to abolish anti-immobility effect of LZ, thereby ruling out the possibility of LZ action through 5-HT_3 receptors. This finding is in accordance with previous similar reports. [33, 34] To study involvement of noradrenergic system in action of LZ, α_1 and α_2-adrenoceptor antagonists were used, namely prazosin and yohimbine respectively. Pretreatment of mice with both these drugs blocked the anti-immobility effect of LZ, indicating that antidepressant-like action of LZ is also mediated by noradrenergic pathway. This finding is in concurrence with reports from previous similar studies where prazosin and yohimbine reversed the action of study compounds. [21, 23, 28] Along with monoaminergic system, another pathway mediating the antidepressant action that has gained importance in the recent years, is the opioid system. An impairment in endogenous opioid system is believed to play a role in the development of depression. [35, 36] Classical antidepressants like tricyclic antidepressants and reuptake inhibitors have been shown to act through opioid receptors. [37-39] Furthermore, a number of compounds tested for antidepressant-like activity have shown to act by modulating

the opioid system. [5, 6, 40, 41] In the current study, pretreatment with naloxone abolished anti-immobility effect of LZ in FST, thereby indicating an action of LZ through opioid receptors. This finding is in concurrence with above-mentioned previous studies. In addition to the elucidated mechanisms, there may be other pathways, through modulation of which, antidepressant-like action of lutein-zeaxanthin may be brought about. These were not studied in the present study. Antioxidant capacity of xanthophylls studied (lutein and zeaxanthin) may be responsible for neuroprotective effect. Brain-derived neurotrophic factor (BDNF) is an important modulator of synaptic plasticity and cell growth, especially formation and survival of serotonin neurons, and it helps in innervation of various regions in the brain. It continues to influence the ability of serotonin system to adapt to various stimuli throughout life. [42] A recent study observed that lutein is capable of increasing serum BDNF levels in humans, while at the same time it was observed to prevent diabetes-induced depletion of BDNF in animals. [43-46] This increase in serum BDNF levels may also contribute towards neuroprotection and antidepressant-like action of LZ, though it was not studied in the present study. Opioid system involvement in LZ action needs to be further investigated, regarding specific opioid receptors involved.

Conclusion:

Results of the present study indicate an antidepressant-like action of lutein-zeaxanthin on acute and chronic oral administration in animal behaviour despair models. In addition, the study also provides evidence for LZ action mediated by serotonergic (5-$HT_{1A/1B}$ and 5-$HT_{2A/2C}$ receptors, but not 5-HT_3 receptors), noradrenergic (α_1 and α_2 receptors) and opioid pathways. Data does contribute to a better understanding of the role played by xanthophylls in CNS, indicating that it could constitute a new and attractive strategy for management of depression. However, future experimental and clinical trials may be needed to determine whether lutein-zeaxanthin will replicate antidepressant-like effect in human subjects suffering from depression.

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