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Acute light deprivation triggers anxiety-like and depressive behaviors in mice

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Abstract:

Light exposure plays a critical role in regulating circadian rhythms and mood-related behaviors. While chronic, constant light deprivation has been extensively studied pertaining to anxiety and depression, the impact of acute deprivation of light exposure remains poorly understood. This study investigated the acute light deprivation-induced effects on anxiety and depressive-like behaviors of male C57BL/6 mice. Six behavioral tests were employed to comprehensively assess anxiety-like and depressive-like levels, including the open field test (OFT), elevated zero maze (EZM), light-dark box test (LDT), sucrose splash test (SST), forced swim test (FST), and tail suspension test (TST). The results indicated that there was no significant difference in anxiety-related measures between the control and light-deprived mice. In contrast, depressive-like behaviors were significantly elevated in the light-deprived mice, as evidenced by reduced self-grooming behavior in the SST and increased immobility in stress-inducing environments in the FST and TST. These findings suggest that even short-term and moderate light deprivation can induce depressive phenotypes, emphasizing the sensitivity of mood regulation to environmental changes, especially light exposure, and highlighting the potential mental health risks faced by urban populations with restricted access to natural light. Further studies are warranted to elucidate the underlying neurobiological mechanisms.

Keywords: Light deprivation; Anxiety; Depression; Seasonal affective disorder; Mental health

1 Introduction

Depression and anxiety represent a significant global health challenge. According to the World Health Organization, over 322 million people worldwide suffered from depression and over 264 million people suffered from anxiety in 2015, with prevalence rates continuing to rise [1]. These disorders frequently co-occur, with nearly 50% of individuals diagnosed with depression also experiencing anxiety [2]. Together, they impose a staggering societal burden, costing over $1 trillion annually in lost productivity

linked to absenteeism and impaired work performance [3]. While numerous environmental factors regulate mood, emerging evidence highlights light exposure as a critical regulator of physiological and psychological processes in both humans [4] and animal models [5]. It is noteworthy that, light availability—or its absence—directly influences anxiety and depression levels.

Animal studies reveal that chronic light deprivation induces mood disturbances. For example, Welberg (2008) reported that three and five weeks of constant darkness in mice resulted in anhedonia, and increased immobility in the forced swimming test and tail suspension test, indicating a strong link between prolonged light deprivation and depressive behaviors [6]. While some studies report elevated anxiety-like behaviors under chronic light deprivation [7], others found no significant changes [8], suggesting depressive symptoms may dominate in prolonged darkness.

The mechanisms underlying these effects are increasingly understood. Chronic light deprivation damages monoamine neurons (noradrenergic, serotonergic, and dopaminergic)—key systems for mood stability—by triggering apoptosis [9]. This neuronal loss correlates with depressive and anxious phenotypes in rodents. Furthermore, the circadian system, critical for regulating sleep, hormonal balance, and mood, becomes dysregulated without consistent light exposure, exacerbating mood disturbances [10]. At the molecular level, altered light conditions disrupt proteins such as HINT1, activating apoptotic pathways that impair both mood and cognition [8]. Despite these insights, critical gaps persist. Existing research predominantly focuses on extreme cases, such as chronic constant darkness or seasonal affective disorder (SAD), a depressive subtype tied to reduced winter daylight in high-latitude regions [11], while urban populations experiencing moderate light insufficiency remain understudied [12]. Emerging evidence indicates insufficient light exposure is not confined to seasonal or geographic extremes: an epidemiological survey found that individuals with inadequate natural light exposure face a 1.5-fold higher risk of depression, a growing concern in modern indoor-centric societies [14]. Moreover, while chronic deprivation is well-documented, the impacts of acute and short-term light reduction remain poorly characterized, despite their potential relevance to urban mental health.

Our study addresses this gap by investigating acute light deprivation, defined as a four-hour daily reduction in light exposure over one week. Unlike prior research emphasizing complete darkness or prolonged deprivation, we examine whether moderate, transient light reductions suffice to induce depression- and anxiety-like behaviors. Given light's role in circadian regulation, we hypothesize that acute deprivation will preferentially disrupt mood, manifesting as depressive symptoms with minimal anxiety effects. We revealed acute light deprivation-induced depressive phenotypes, including reduced sucrose preference and increased immobility in forced swim and tail suspension tests, whereas anxiety-like behaviors remained unchanged. These findings challenge the notion that only extreme or prolonged light deprivation impacts mental health, underscoring the sensitivity of mood regulation to even transient light reductions.

This work highlights the mental health risks of acute light insufficiency, particularly for urban populations with limited natural light access. However, by focusing solely on behavioral outcomes, this study leaves underlying neurobiological mechanisms unexplored. Future research should investigate neurotransmitter dynamics, neuroinflammatory pathways, and circadian gene expression to elucidate the biological basis of light deprivation's effects. Such insights could inform public health strategies to mitigate mood disorders in increasingly indoor-oriented societies.

2 Materials and Methods

2.1 Animals and light deprivation

14 adult C57BL/6 male mice, 8 weeks old, were purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd., Beijing, China, and housed under standard laboratory conditions with ad libitum access to food and water. Animals were treated ethically by adhering to the international guidelines on the ethical use of experimental animals.

Mice were randomly assigned to two groups: the light deprivation (LD) group (n=7) and the control group (n=7). The mice of the light deprivation group were subjected to an additional 4 hours of darkness per day for 7 days. This meant that the dark phase of the light cycle was extended to 16 hr/day while the light phase was reduced to 8 hr/day. The mice of the control group remained on a 12-hour/12-hourlight/dark cycle.

2.2 Behavioral tests

Six behavioral tests were conducted to evaluate the effects of acute light deprivation. The behavior of mice in the test was recorded using a digital camera. The videos of all behavioral tests, except the open field test, were analyzed by an experimenter who was blind to the experiment. The apparatuses used were cleaned with 75% ethanol between trials to avoid any odor cues.

2.2.1 Open field test

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The open field test (OFT) was used to assess locomotor activity and exploratory behavior in order to monitor anxiety-like behavior [14]. The OFT apparatus consisted of a brightly illuminated 40 × 40 cm square arena surrounded by a 40 cm-high wall. Mice were individually placed in the center of a clear and unfamiliar 40 cm x 40 cm area with 30 cm high walls to minimize external visual distraction. The center zone was defined as a square area located in the middle of the arena, measuring 20 cm x 20 cm, which is half of the total width and half of the total length. The test was recorded for 5 minutes, and behavioral data were analyzed using the tracking software EthoVision XT 16, which extracts parameters including the total distance traveled, time in the center area, distance traveled in the center area, and the number of center area entries. Avoidance of the center zone and reduced center exploration were indicators of increased anxiety-like behavior, whereas increased center exploration suggested lower anxiety levels [15].

2.2.2 Elevated zero maze

The anxiety-like behavior of mice was also analyzed using the elevated zero maze (EZM) by evaluating the exploratory activity under risks. The maze consisted of a 50 cm diameter circular platform elevated 60 cm above the floor and was divided into four equal-sized sections: two opposing open sections and two opposing enclosed sections. The enclosed sections were surrounded by 15 cm high opaque walls, whereas the open sections had no walls, exposing the mice to potential risks. Mice were placed on the border of an open and an enclosed section facing the closed quadrant. This test lasted for 5 min. The time that mice spent in the open arms, the number of entries into the open arms, and the latency to the first entry into the open arms were analyzed manually. An entry into the open arm was defined as half of the body crossing the boundary into the open section. A reduction in open-section exploration and entry times was considered indicative of heightened anxiety levels [16].

2.2.3 Light-dark box test

The light-dark box test (LDT) was conducted to evaluate anxiety-like behavior by measuring the conflict between the rodent's natural preference for dark, enclosed spaces and its innate tendency to explore new environments [17]. The apparatus consisted of a 45 cm x 27 cm x 30 cm two-chambered box; one-third of the total area was a dark compartment, fully enclosed with black-colored walls, while the remaining two-thirds was a brightly lit compartment. A 15 cm x 10 cm opening connected the two compartments, allowing the mice to move freely between them. Mice were placed at the center of the light box with their back to the dark area. The test is 10 minutes. The behavioral parameters recorded and analyzed included the total time spent in the light area, the number of light area entries, and the latency to first enter the dark area. An entry into a compartment was defined as half of the body crossing the boundary between the two chambers. Increased anxiety-like behavior was indicated by spending less time in the light compartment, a longer latency to enter the light area, fewer transitions between compartments, and a shorter latency to enter the dark area [18].

2.2.4 Sucrose splash test

The Sucrose Splash Test (SST) was used to evaluate anhedonia, a core symptom of depression, by measuring self-care and grooming behavior in response to sucrose application [19]. The test is based on the principle that sucrose has a rewarding and pleasurable effect on mice, and a lack of motivation to engage in grooming after sucrose application shows reduced hedonic drive and motivational deficits associated with depression-like states. At the start of the test, a 10% sucrose solution was sprayed onto the mice's dorsal fur. The sucrose creates a mildly sticky sensation to trigger grooming behaviors, and as rodents find sucrose rewarding, they will keep licking and grooming themselves to remove it. Immediately after sucrose application, each mouse was placed in an individual testing cage, and the behavior of the mice was evaluated for 5 minutes to quantify total grooming time and the number of groomings. A decrease in grooming behavior was interpreted as a sign of anhedonia and depression [20].

2.2.5 Forced swimming test

The forced swimming test (FST) could assess behavioral despair and depressive-like behaviors in rodents by evaluating their response when placed in an inescapable stressful environment [21]. Mice are individually placed in a cylindrical tank 24 cm in height and 10 cm in diameter, filled with 23 ±2°C water. The water depth was 16 cm, which was sufficient to prevent the mice from touching the bottom while allowing free movement. The experiment lasted for 6 min, and the duration of mobility was scored. Mobility included swimming, which involved horizontal movement through the water, and struggling or climbing, which consisted of vigorous attempts to escape the tank. Periods where the mice floated passively and lacked any active swimming movements, making only minimal movements necessary to keep their head above water, were classified as immobility and were excluded from the mobility time calculation. A lower mobility time suggested a depression-like state. The rationale behind this is that giving up escape behaviors early after initial attempts failed reflects learned helplessness, a key feature

of depression-like behavior in animal models [22].

2.2.6 Tail suspension test

Similar to FST, the tail suspension test (TST) was performed to evaluate behavioral despair and learned helplessness, which were indices of depression after exposure to stress stimuli [23]. Each mouse was securely suspended by its tail using adhesive tapes attached to a hook 30cm above the floor. Handling the rats should be conducted as quickly as possible with gentle movements to minimize the excess stress and stimuli to the rats. The video should record the full body movements of rats. The total duration of the experiment is 6 minutes and the total time of mobility, latency to first immobility, and number of mobilities were assessed. Mobility was defined as the mouse actively moving its limbs, swinging its body, and attempting to climb up its tail to escape. A shorter mobility time correlates with behavioral despair, a key symptom of depression [24].

2.3 Statistical analysis:

The statistical differences were analyzed through GraphPad Prism software using the t-tests, and the significant difference was set at $p<0.05$.

3 Results

3.1 Acute light deprivation did not influence anxiety-like behaviors

To evaluate the effects of acute light deprivation on anxiety-related behaviors, three behavioral tests were conducted: the open field test (OFT), elevated zero maze (EZM), and light-dark box (LDB) test. These tests assess different aspects of anxiety-related behavior by measuring an animal's willingness to explore aversive environments, with a reduced exploration of open or brightly lit spaces typically interpreted as increased anxiety-like behavior.

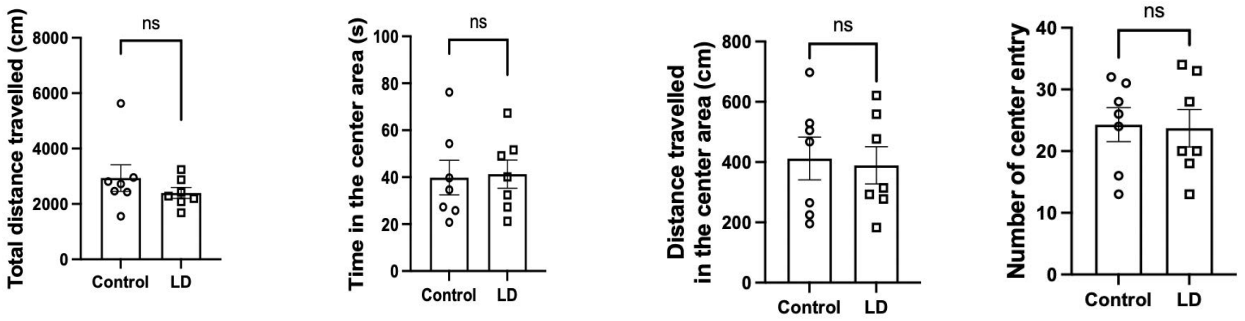

Figure 1. Acute light deprivation did not affect anxiety-like behavior in OFT.

In the OFT, the time spent in the center, the distance traveled in the center, and the number of center entries by LD and control groups are almost identical, as illustrated in Figure 1. There is no significant decrease in center exploration; hence, no difference in anxiety levels. The total distance traveled, which measures locomotor activity, was slightly lower in the LD group, but the difference was not statistically significant. This result is more likely attributed to general variability in movement rather than anxiety-related alterations. These findings suggest that acute light deprivation did not significantly impact the exploratory behavior of mice in the OFT, providing little evidence for an increase in anxiety-like behavior.

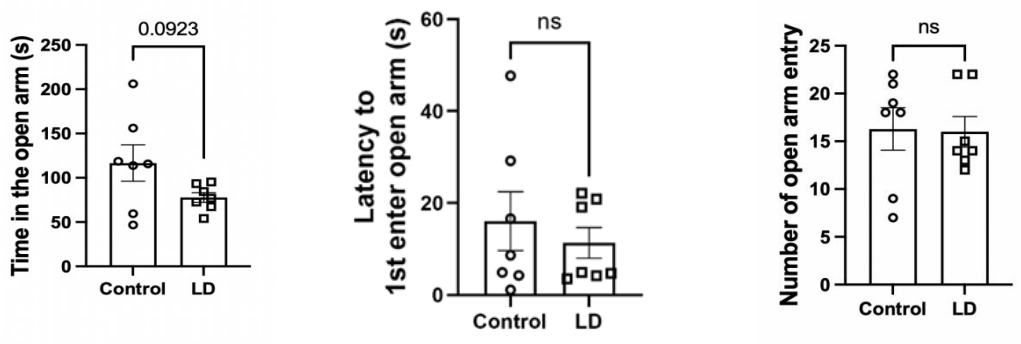

Figure 2. Mice showed increased anxiety-like behavior in the EZM after acute light deprivation.

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In the EZM test, the LD group spent less time in the open arms, but the difference was not statistically significant (Figure 2). The latency to the first entry into the open arm and the number of open arm entries showed no significant differences between groups. These results in the EZM indicate that LD groups failed to show anxiety-like behaviors caused by acute light deprivation.

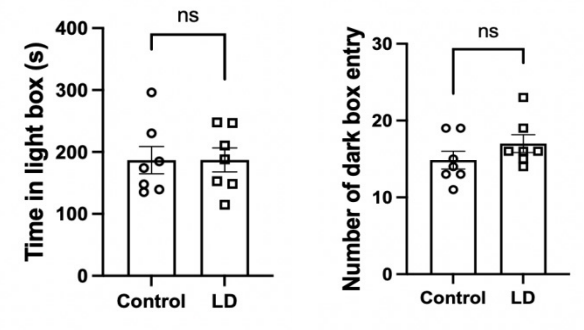

Figure 3. Acute light deprivation did not affect anxiety-like behavior in LDT.

The mice again showed no significant differences in the time spent in the light box and the number of light box entries in LDT as shown in Figure 3. It indicates that the LD mice and control mice have a similar tendency to avoid the bright environment, reflecting the same risk aversion. Together, acute light deprivation does not alter anxiety behaviors in mice.

3.2 Acute light deprivation intensifies depression-like behaviors

Next, we want to evaluate whether acute light deprivation could change depression-like behavior. The sucrose splash test (SST), forced-swim test (FST), and tail-suspension test (TST) were performed on the control mice and LD mice.

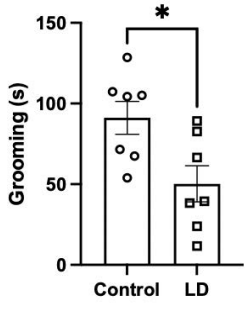

Figure 4. The LD mice showed a decline in grooming time in SST compared to control mice.

In the SST, the LD group exhibited a significant reduction in grooming time compared to the control group, interpreted as anhedonia, a main symptom of depression, suggesting a decrease in self-care and motivational behaviors as shown in Figure 4.

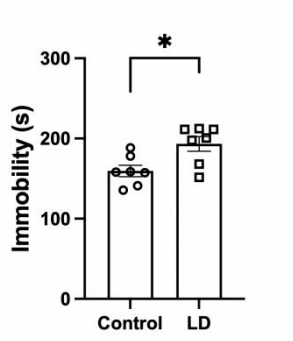

Figure 5. The LD mice showed enhanced immobility time in FST compared to control mice.

In the FST, Figure 5 shows that there's a significant increase in immobility time between the LD group and the control group, indicating a greater tendency toward behavioral despair, suggesting that control mice attempted to escape for a longer duration before adopting a passive state, whereas LD mice gave up more quickly.

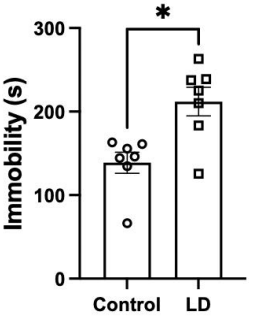

Figure 6. The LD mice showed increased immobility time in TST compared to control mice.

The result of TST in Figure 6 is consistent with the FST. The LD group exhibited significantly longer immobility times, suggesting that LD mice rapidly adopted a passive posture, again proving a higher level of depression and learned helplessness. These results indicate that acute light deprivation could induce despair in mice.

4 Discussion

This study investigated the effects of acute light deprivation (LD) on mood disorders, revealing a significant

increase in depressive-like behaviors but no substantial effect on anxiety-like behaviors. The LD group displayed reduced grooming time in the SST, and increased immobility time in FST and TST, suggesting that even a reduction in daily light exposure can negatively impact mood regulation. However, the absence of significant changes in most anxiety-related parameters for the OFT, EZM, and LDT indicates that acute light deprivation does not affect anxiety-like behavior at all. These results support the hypothesis that acute light deprivation primarily induces depressive behaviors while exerting little effect on anxiety.

The findings of this study are consistent with the well-established association between light exposure and mood regulation. Light has been shown to influence mood and cognitive functions through intrinsically photosensitive retinal ganglion cells, which affect both the circadian system and mood-regulating brain regions. Thus, lack of light has a direct negative impact on mood [25]. The absence of significant anxiety-related behavioral changes in this study also aligns with previous research by Zhou et al. [8]. A key contrast between this study and prior work is the duration of light deprivation required to induce depressive-like behaviors. Previous studies have suggested that chronic light deprivation, often lasting a minimum of three weeks in complete 24-hour darkness, is necessary to exhibit significant depression-like symptoms in rodents [26, 27]. However, the present study demonstrates that even an acute 4-hour reduction in daily light exposure with a total 16-hour darkness over a shorter one-week period is sufficient to induce depressive-like behaviors.

The current work introduces a novel perspective by demonstrating that acute, moderate reductions in light exposure can induce depressive-like behaviors. This challenges traditional models that associate depression-like behaviors only with chronic, constant light deprivation. The use of multiple behavioral tests for depression and anxiety strengthens the validity of the findings, providing a more comprehensive assessment of the effects of acute light deprivation on mood-related behaviors. However, there are several limitations. The study relies solely on behavioral assessments, leaving the underlying cellular and molecular mechanisms unexplored. Future research should investigate different neural pathways specifically altered in the circadian gene expression [28], serotonin and dopamine signaling [29], and neuroinflammation [30], which are proven to be linked with the occurrence of depression. Additionally, this study only examined rats' behaviors once, which makes it unclear whether the depressive-like behaviors persist over time to show major depression. Major depression in rodent models is typically defined by at least one core symptom persisting for a minimum of two weeks [31]. Overall, this finding of acute light deprivation's effects on mood has broad implications for public health, as it suggests that individuals living in environments with reduced natural light, such as urban populations, may be at an underrecognized risk for mood disturbances, especially depression.

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Supplementary:

Table 1. Anxiety-like behavioral tests

		Control	Light deprivation
OFT	Total distance traveled (cm)	2938.42	2398.30
	Time in the center area (s)	39.82	41.30
	Distance traveled in the center (cm)	411.97	389.21
	Number of center area entry	24.29	23.71
EZM	Time in open arm (s)	116.66	77.71
	Latency to 1st into the open arm (s)	16.07	11.36
	Number of open arm entry (s)	16.29	16.00
LDT	Time in light box (s)	186.77	187.04
	Number of light box entry	14.86	17.00
	Latency to 1st enter dark box (s)	14.06	12.89

Table 2. Depressive-like behavioral tests

		Control	Light deprivation
SST	Time of grooming (s)	91.14	50.24
	Number of grooming	25.43	15.57
FST	Immobility time (s)	159.56	193.30
	Latency to 1st into immobility (s)	67.00	54.87
	Number of mobility	18.43	21.29
TST	Immobility time (s)	138.79	211.96
	Latency to 1st into immobility (s)	35.14	7.54
	Number of mobility	19.29	29.29