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The modulatory role of 6-gingerol-rich portion of ginger on brain oxidative stress, inflammation and neurotransmission in chronic unpredicted mild stress-induced depressed male Wistar rats

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Abstract

Objective: This study was designed to investigate the effect of 6-gingerol-rich extract of ginger (6-GIRPOG) on oxidative stress, inflammation and neurotransmission in chronic unpredicted mild stress (CUMS)-induced depressed male Wistar rats.

Materials and Methods: Twenty-five (25) male Wistar rats in total were divided into five groups at random (n = 5 in each group). The control group received 0.5 ml of normal saline, CUMS rats were only exposed to CUMS daily, CUMS + fluoxetine rats were exposed to CUMS and orally received 10 mg/kg per body weight of fluoxetine daily, CUMS + 6-GIRPOG (100) and CUMS + 6-GIRPOG (200) rats were exposed to CUMS and orally received 100 and 200 mg/kg body weight of 6-GIRPOG respectively (daily). Exposure to CUMS and treatment were carried out for a period of 21 days, after which light and dark box test, sucrose splash test, and forced swim test were conducted to assess the behavioral functions. The rats were then euthanized and their brain samples were collected for biochemical analysis.

Results: The exposure to CUMS caused behavioral alterations as well as a significant ($p<0.05$) decrease in the levels of brain-derived neurotrophic factor (BDNF), glutamate, reduced glutathione (GSH), and activities of superoxide dismutase (SOD). Furthermore, the exposure to CUMS caused a statistically significant rise ($p<0.05$) in the brain norepinephrine, cortisol, nuclear factor kappa-B (NFK-B), tumor necrosis factor-alpha (TNF-α), interleukin-1 beta (IL-1β), and IL-6. However, the administration of both 100 and 200 mg/kg of 6-GIRPOG effectively reversed these behavioral and biochemical changes.

Conclusion: Consequently, the study reveals the role of 6-GIRPOG in ameliorating CUMS-induced depression and brain damage via antioxidative, anti-inflammatory, and neurotransmission modulatory mechanisms.

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Introduction

Depression has a significant negative influence on the quality of life and cause significant financial costs for both individuals and society (WHO 2021). With high rates of suicide, recurrence, and prevalence, it is currently the most common type of psychiatric condition. It was reported that 322 million people worldwide suffer from depression, and that its prevalence is 4.4% (WHO 2017). Stress-induced depression causes intricate changes to the nervous system. Numerous studies have shown a connection between the hypothalamic-pituitary-adrenal (HPA) axis dysfunction and the neuropathology of major depression (Bai et al. 2018). Patients with significant depression have higher serum cortisol concentrations as a result of the hyperactivation of the HPA axis (Keller et al. 2017). One important neuroendocrine system that controls the bio-behavioral reaction to stress is the HPA axis. The adrenal glands produce the glucocorticoid hormone cortisol, which is essential to many bodily physiological functions (Knezevic et al. 2023). Since their discovery and use in clinical settings decades ago, glucocorticoids have been known to have a potent anti-inflammatory impact. Contrarily, exposure to long-term stress induces the slow emergence of glucocorticoid resistance, which in turn, fuels persistent, irreversible inflammation in the central nervous system (Knezevic et al. 2023). The HPA axis and sympathetic nervous system (SNS) are triggered by social conflicts and stress. Norepinephrine (NE), which is released when the two work together, increases the transcription of the proinflammatory immune response genes tumor necrosis factor-alpha(TNF-α), interleukin-1 beta (IL-1β), and interleukin-6 (IL-6) (Lee et al. 2015). Another mechanism involved in the neurobiology of depression is oxidative stress (Vaváková et al. 2015). Therefore, it has been thought that treating depression may involve reducing oxidative stress and strengthening the body's defenses against oxidative damage.

The term chronic unpredictable mild stress (CUMS) refers to the study of neurobiological alterations brought on by prolonged exposure to psychological stress (Dionisie 2024). CUMS primary goal is to induce behavioral abnormalities in mice and rats, such as anhedonia and behavioral despair, in order to facilitate the search for possible medicinal pharmaceuticals (Burstei et al. 2018). Lopez-López et al. reported that CUMS is the application of different treatments as stressors that are comparable to regular stress but not too intense and are administered for a minimum of three weeks (Lopez-Lopez et al. 2016). Wider alterations and adaptations can result from repetitive stress, particularly when it comes to modifications in energy metabolism (Koch et al. 2023).

Even with the wide range of antidepressants available, treating depression still requires overcoming a few challenges. It is necessary to look for safer and more effective antidepressants due to inefficacy, adverse effects, treatment resistance, and the delayed onset of action for some antidepressants (Rafeyan et al. 2020). For psychiatric problems, medicinal plants are typically suggested as an alternative treatment. Numerous bioactive substances found in medicinal plants have the ability to both lessen and enhance one another's therapeutic benefits (Gavzan et al. 2023). There have been reports of the ability of *Zingiber officinale*, also known as ginger, to combat ulcer, inflammation, tumor, oxidative stress, digestive and gastric problems. In Chinese traditional medicine, it is utilized as interventions for a variety of gastrointestinal and respiratory conditions as a stomachic, antiemetic, antidiarrheal, and cardiotonic. Motion sickness can also be treated with powdered ginger (Shalinee et al. 2022). It is clear from the literature that ginger may both shield neurons and decrease depressive-like activities in animals (Benkermiche et al. 2022). Although there have been a few

publications of ginger's antidepressant properties, none have discussed the potential of 6-gingerol to treat depression (Phukan and Adhikari 2017; Kukula-Koch et al. 2018). Therefore, the aim of this research is to find out the role of 6-gingerol-rich portion of ginger in chronic unpredicted mild stress (CUMS)-induced depression in male Wistar rats.

Materials and Methods

Rats

From the animal house of the Biochemistry Department, Faculty of Life Sciences, University of Ilorin, Ilorin, Nigeria, twenty-five (25) male Wistar rats weighing between 150 and 200 g were acquired. The rats were acclimated to the surroundings for 14 days prior to the start of the investigation. The University of Ilorin's Ethical Review Committee approved all of the experiment's procedures; the approval number was UERC/ASN/2023/2451, and the study was carried out in compliance with the Helsinki Declaration and the International Principles governing research on animals.

Drugs, chemicals and reagents

Mich-Mikedenson Laboratory, Taiwo Road, Ilorin, Nigeria, provided the ketamine (Parkie-Davies, Freiburg, Germany), while Mamrota Pharmacy, Ilorin, Nigeria, provided the fluoxetine (Chemo-pharma Laboratories Ltd., Lagos, Nigeria).

Ginger rhizomes

Ginger rhizomes were procured from Ipata market in Kwara State, Nigeria. The voucher number UILH/001/1168/2022 was issued when the rhizomes were identified and confirmed by the Department of Plant Biology, Faculty of Life Sciences, University of Ilorin.

Preparation and isolation of 6-gingerol-rich portion of Ginger (6-GIRPOG)

After being rinsed with clean water, chopping and slicing into pieces, air-drying, and crushing of the ginger rhizomes into powdery form were carried out. Also, extraction was done for 3 days using 95% ethanol, after which isolation was carried out (Asuku et al. 2024).

High performance liquid chromatography

The samples were subjected to high performance liquid chromatography (HPLC) in order to accomplish component separation, identification, and quantification. The method of Asuku et al. was employed for the HPLC. The 6-GIRPOG samples were collected for HPLC analysis into a 25 ml standard flask after being diluted with acetonitrile and water. Ubondapak (C-18 column) served as the stationary phase. Water and acetonitrile were used as the mobile phase, and the flow rate was 1 mL per minute. Pump A and B carried water and acetonitrile respectively. A rheodyne injector (10 µl loop) was used to inject the sample. The separation into different constituents was performed at room temperature. The samples were subjected to a diode array detector (254 nm) following column separation, which allowed for the identification as well as quantification of each individual constituent (Asuku et al. 2024). Table 1 lists the 14 active constituents in 6-GIRPOG, with 6-gingerol having the highest percentage (51.05%).

Table 1. HPLC identification and quantification of 6-GIRPOG components

Components	Percentage (%)
Sinapinic Acid	8.4
P- Coumaric Acid	18.23
Benzoic Acid	5.05
Alpha Farnesen	1.96
Caffeic Acid	1.03
Zingiberene	0.74
Gingerenone-A	0.82
Cinnamic Acid	0.88
6-Gingerol	51.05
Zingerone	6.31
Apigenidin	0.76
Syringic Acid	3.19
Ferulic Acid	0.77
Betaine	0.67

Chronic unpredictable mild stress (CUMS)

Over the course of three weeks, the rats were continuously exposed to a variety of low-intensity, randomly scheduled social and environmental stressors. Overcrowding, isolation, denial of food and water, cage tilting (45 degrees), wet bedding (200 ml of water per cage), sawdust removal, and cage wetting (200 ml of water per cage) are some of these stressors. One of these stressors was given to rats every day. In order to prevent the animals from anticipating when they would be stimulated, the same stressor was not applied for three days in a row. Body weight was measured before exposure to the stressor and after exposure and drugs administration.

Experimental design

The investigation was conducted in Ilorin, Nigeria, at the Bioresearch Hub Laboratory Limited. A total of twenty-five (25) male Wistar rats were randomly assigned to five groups (n = 5 in each group). To the control group, 0.5 ml of normal saline was given orally. CUMS rats were only exposed to CUMS daily, CUMS + fluoxetine rats were exposed to CUMS and orally received 10 mg/kg per body weight of fluoxetine daily, CUMS + 6-GIRPOG (100) and CUMS + 6-GIRPOG (200) rats were exposed to CUMS and orally received 100 and 200 mg/kg body weight of 6-GIRPOG respectively (daily). Exposure to CUMS and treatment were carried out for a period of 21 days. Table 2 shows the groupings of the rats and the administered dosages of drugs.

Table 2. Groupings of the rats and the administered dosages of drugs

	Group (n=5)	Treatments
1.	Control	Rats received orally administered normal saline (0.5 m).
2.	CUMS only	Rats were only exposed to CUMS daily
3.	CUMS + Fluoxetine	Rats were exposed to CUMS and orally received 10 mg/kg per body weight of fluoxetine daily
4.	CUMS + 6-GIRPOG (100)	Rats were exposed to CUMS and orally received 100 mg/kg body weight of 6-GIRPOG daily (Asuku et al. 2024)
5.	CUMS + 6-GIRPOG (200)	Rats were exposed to CUMS and orally received 200 mg/kg body weight of 6-GIRPOG daily (Asuku et al. 2024)

CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich extract of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich extract of ginger (200 mg/kg body weight).

Determination of behavioral functions

Before the exposure of rats to CUMS and administration of drugs, there was baseline behavioral training, and after the exposure and administration, behavioral tests (light and dark box test, sucrose splash test, and forced swim test) were carried out on the animals on day 21.

Light and dark box

A well-liked animal model in pharmacology for testing rats' unconditioned anxiety response is the light and dark box test. There are two compartments in the light and dark box apparatus. The small dark compartment, which makes up 1/3 of the box, is covered and gloomy, while the huge light compartment, which makes up 2/3 of the box, is well-lit and open. The two chambers are linked by a 7 cm door. The rat was left free to roam between the two chambers and was positioned in the center of the well-lit space. Animal behaviors were captured using the webcam recorder, and the amount of time the animals spent in the light/dark compartment was examined.

Sucrose splash test

This test was run with a 15 W red light on. Rats in their home cage were given a 10% sucrose solution via an atomizer spray applied to their dorsal coat. The coat gets dirty from the sucrose solution, and grooming behavior is induced. As a measure of self-care and motivating behavior, the amount of time spent grooming and grooming latency were monitored for five minutes following the application of the sucrose solution.

Forced swim test

A glass cylinder measuring 25 cm in height and 10 cm in diameter was filled with water at a temperature of 22–25°C until it reached a height of 10 cm for each rat. During the test, each rat was given five minutes to swim, and in the last four minutes, the length of immobility was recorded and observed. The immobility period was the duration rat spent floating in the water without exerting itself and moving only as much as was required to keep its head above the surface. After being dried with a towel, the animals were put back in their enclosures.

Biochemical analysis

After 21 days of treatment, the rats were anesthetized with an intraperitoneal dose of ketamine (75 mg/kg; Asuku et al. 2024). Following their removal on ice, the brain tissues were promptly cleaned with 0.1 molar of phosphate buffer solution (PBS) and stored in the same solution. Five milliliters of ice-cold PBS were used to homogenize brain samples (Haq and Almaro 2018). Following ten minutes of centrifugation at 3000 revolutions per minute, supernatants were collected and used for biochemical analysis. The following were measured using ELISA kit (Elabscience Biotechnology Inc., Texas) in accordance with the guidelines provided by the kit's manufacturer. The superoxide dismutase (SOD) activity, reduced glutathione (GSH) level, cortisol, norepinephrine, glutamate, brain-derived neurotrophic factor (BDNF), interleukin 1-beta (IL-1β), IL-6, tumor necrosis factor-alpha (TNF-α), and nuclear factor kappa-B (NFK-B) levels were measured**.**

Statistical analysis

A one-way ANOVA and Newman Keul's *post hoc* test were used to analyze the data, which were all provided as mean±S.E.M. Analysis was carried out using graph pad prism 9.0. At $p<0.05$, values are statistically significant.

Results

Figure 1 shows the initial weights of the rats (before exposure to CUMS and treatment) and their final weights (after exposure to CUMS and treatment). There was a significant reduction from initial to final weight of the CUMS rats, while there was a significant increase from initial to final weight of the control, fluoxetine and GIRPOG-treated rats.

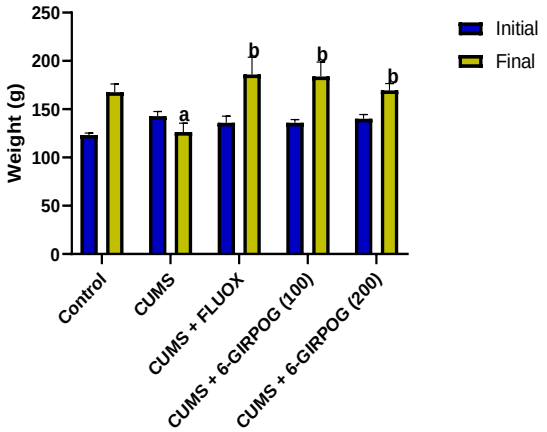

Figure 1. The effect of 6-GIRPOG on the weight of chronic unpredicted mild stress Wistar rats. Data are presented as Mean ± SEM, n = 5; $^{a}p<0.05$ vs. control; $^{b}p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich fraction portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

There was a significant ($p<0.05$) increase in the time spent in the dark box in CUMS-exposed rats compared to the control as shown in Figure 2A, however

administration of both doses of GIRPOG reversed the trend, and this was in line with the group administered with fluoxetine.

The treatment of 6-GIRPOG considerably increased the grooming time in 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats (Figure 2B).

Figure 2C shows a significant ($p<0.05$) increase in the grooming latency of the rats exposed to CUMS. In contrast, treatment with 6-GIRPOG remarkably ($p<0.05$) decreased the grooming latency in the rats treated with 6-GIRPOG. This was in line with the group administered with fluoxetine.

In Figure 2D, the treatment of 6-GIRPOG considerably ($p<0.05$) reduced the immobility time in 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats, however the immobility time of the CUMS-exposed rats was significantly increased when compared to the control rats. This outcome has a favorable comparison to the standard group.

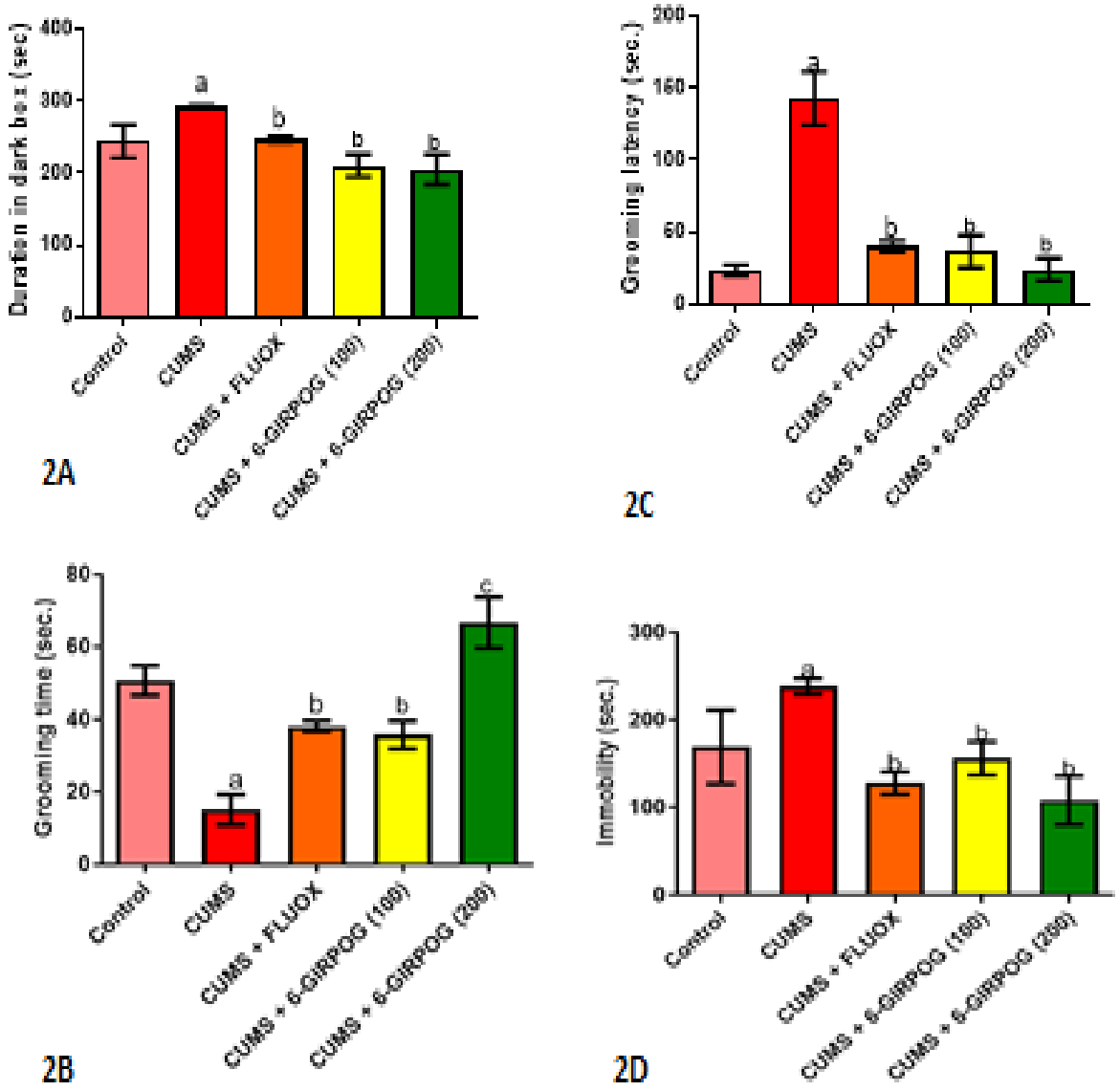

Figure 2. (A, B, C, and D): The effect of 6-GIRPOG on behavioral deficits induced by CUMS. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b, c]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

In Figure 3, exposure to CUMS produced a significant ($p<0.05$) decrease in the BDNF of the CUMS-exposed rats. While the treatment with 6-GIRPOG considerably boosted the BDNF of the 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats.

Figure 4 shows that the brain glutamate level of the CUMS-exposed rats was significantly ($p<0.05$) decreased in comparison to the control rats. On the other hand, 6-GIRPOG significantly ($p<0.05$) raised the brain glutamate level of the rats administered with 6-GIRPOG relative to the CUMS-exposed rats. This outcome was consistent with the fluoxetine group

In Figure 5, the brain norepinephrine level of the CUMS-exposed rats was increased significantly ($p<0.05$) compared to that of the control, but the groups that received 6-GIRPOG had significantly ($p<0.05$) lower brain norepinephrine level than the CUMS-exposed group. This outcome is dose-dependent.

Figure 6 shows that the cortisol levels of the CUMS-exposed rats was increased significantly ($p<0.05$) compared to that of of the control. On the other hand, 6-GIRPOG significantly ($p<0.05$) reduced the cortisol level of the 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats.

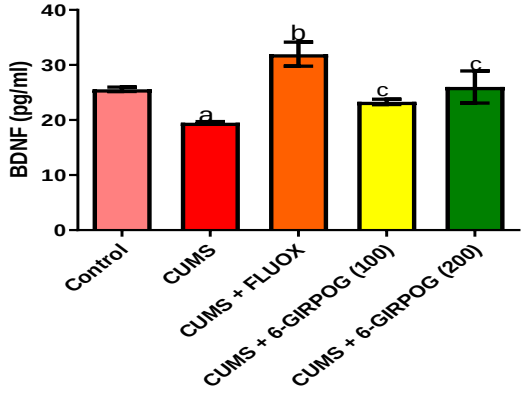

Figure 3. The effect of 6-GIRPOG on brain-derived neurotrophic factor (BDNF) level. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b,c]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

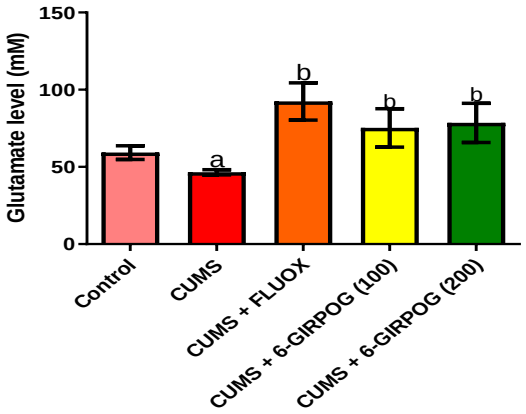

Figure 4. The effect of 6-GIRPOG on glutamate level. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

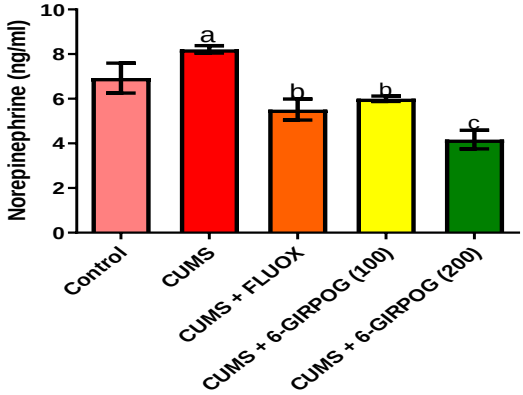

Figure 5. The effect of 6-GIRPOG on norepinephrine level. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b,c]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

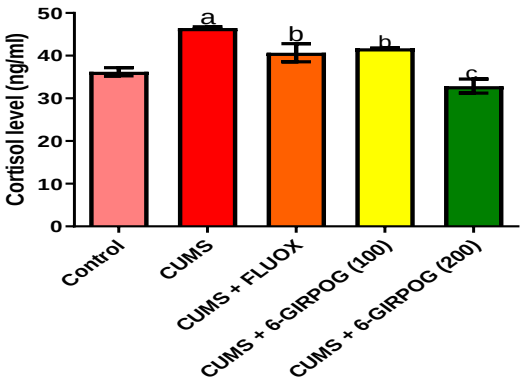

Figure 6. The effect of 6-GIRPOG on cortisol level. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b,c]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

CUMS-exposed rats' brain GSH levels significantly ($p<0.05$) decreased in comparison to the control rats but the treatment with 6-GIRPOG considerably ($p<0.05$) raised the brain GSH levels of the 6-GIRPOG-administered rats in comparison to the CUMS-exposed rats (Figure 7A). This outcome was consistent with the fluoxetine group.

Figure 7B shows that exposure to CUMS brought on a significant ($p<0.05$) decrease in the brain SOD activity of the CUMS-exposed rats in comparison to the control rats while 6-GIRPOG significantly ($p<0.05$) increased the activity of the brain SOD in the 6-GIRPOG-administered groups in comparison to the CUMS-exposed group. There is a dose-dependent rise.

Figure 8A shows that the brain NFK-B level of the CUMS-exposed rats was significantly ($p<0.05$) higher than that of the control. On the other hand, the administration of 6-GIRPOG significantly ($p<0.05$) decreased the brain NFK-B levels of the 6-GIRPOG-treated groups in comparison to the CUMS-exposed group. There is a dose-dependent reduction.

Exposure to CUMS caused a significant ($p<0.05$) increase in the brain TNF-α level of the CUMS-exposed rats in comparison to the control rats, but 6-GIRPOG significantly reduced the brain TNF-α levels of the 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats (Figure 8B).

The exposure to CUMS brought about a significant ($p<0.05$) increase in the brain IL-1β levels of the CUMS-exposed rats in comparison to the control rats, however, the administration of 6-GIRPOG considerably ($p<0.05$) reduced the brain IL-1β levels of the 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats (Figure 8C).

The exposure to CUMS brought about a significant ($p<0.05$) increase in the brain IL-6 level of the CUMS-exposed rats in comparison to the control rats, but the treatment with 6-GIRPOG considerably ($p<0.05$) reduced the brain IL-6 levels of the 6-GIRPOG-treated rats in comparison to the CUMS-exposed rats (Figure 8D). This outcome compares favorably to the group that received fluoxetine treatment.

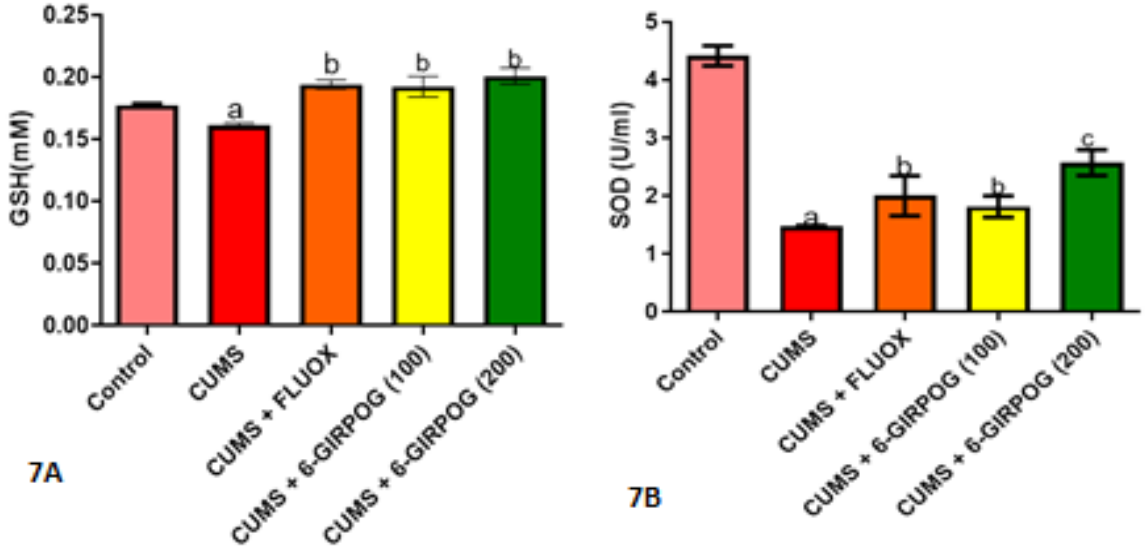

Figure 3. (A and B): The effect of 6-GIRPOG on antioxidants status and activity. Data are presented as Mean ± SEM, n = 5; [a]$p<0.05$ vs. control; [b,c]$p<0.05$ vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

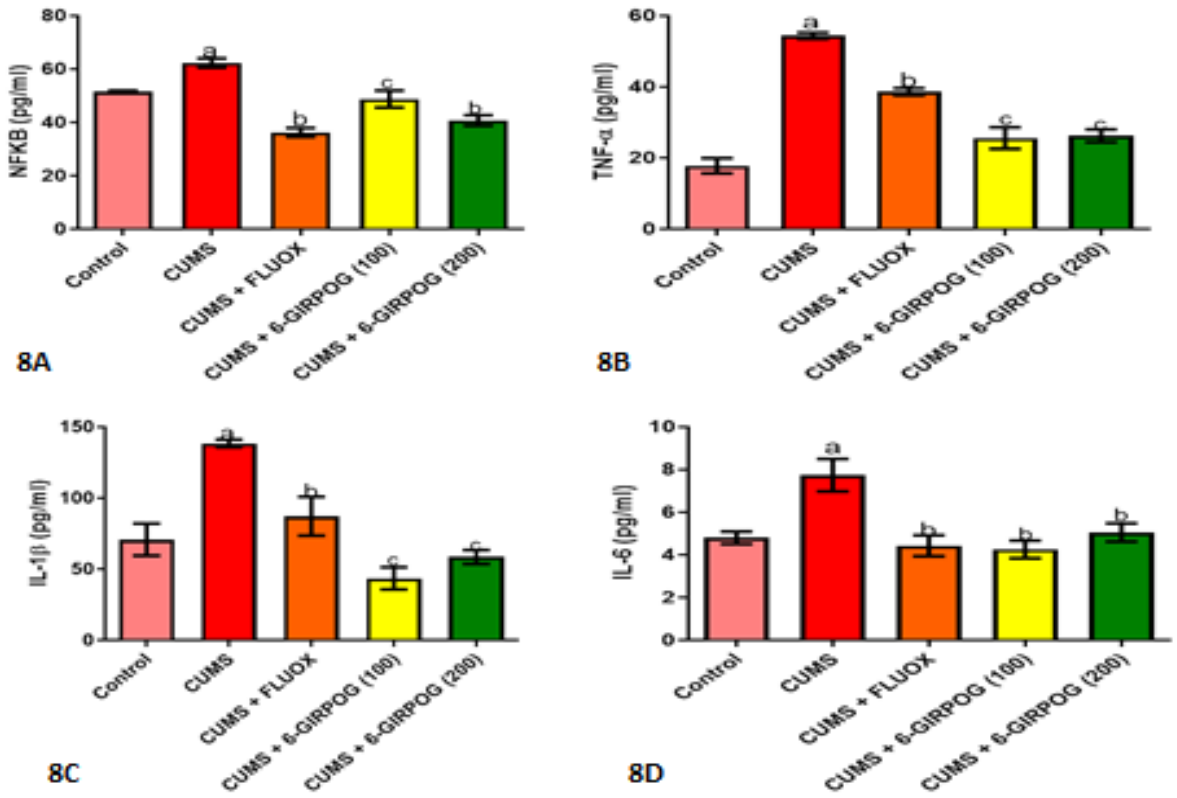

Figure 4. (A, B, C, and D): The effect of 6-GIRPOG on pro-inflammatory markers levels. Data are presented as Mean ± SEM, n = 5; [a]p<0.05 vs. control; [b,c]p<0.05 vs. CUMS. CUMS, Chronic unpredicted mild stress; 6-GIRPOG (100), 6-gingerol-rich portion of ginger (100 mg/kg body weight); 6-GIRPOG (200), 6-gingerol-rich portion of ginger (200 mg/kg body weight); FLUOX, Fluoxetine.

Discussion

In this study, we found that the body weight of CUMS-induced rats significantly decreased, that they spent significantly much time in the dark box during the light and dark box tests, that they significantly reduced their grooming behaviors during the splash test, and that they significantly increased their immobility duration during the forced swim test. This finding corroborates the theory put forth by Kingir et al. that stressful situations can alter behavior (Kingir et al. 2023). The tendency was considerably reversed by administering 6-GIRPOG, which may have been caused by its antidepressant effects. The current findings support earlier research that shown that several medicinal plants have an antidepressant effect on rats with CUMS-induced behavioral impairments (Aryanezhad et al. 2021).

Wei et al. reported that CUMS promotes the activation of the HPA-axis and the secretion of molecules associated to it, including cortisol and corticosterone (Wei et al. 2017). According to the current findings, there was a discernible rise in brain cortisol levels in the CUMS. While fluoxetine and 6-GIRPOG both decreased the level of cortisol, the difference between them and the control group was not statistically significant. Therefore, it is possible to argue that 6-GIRPOG's attack on CUMS does not target the HPA axis. In order to cease responding to stress, cortisol usually provides negative feedback by decreasing Corticotropin releasing hormone secretion. In contrast, the HPA axis is always active during periods of chronic stress. Prolonged stress can cause cortisol levels to rise, which can affect many of the body's normal functions, including weight fluctuations as seen in the CUMS.

It was shown that depression and stress reduce BDNF blood levels and reduce the BDNF expression in brain areas that play part in cognition and mood control (Cavaleri et al. 2023). The relationship between glutamate transmission and BDNF is intricate. Neuronal glutamate sensitivity is altered by BDNF which is produced and released in response to glutamate. Moreover, decreased BDNF expression and release are intimately linked to aberrant glutamate transmission (Murrough et al. 2017). According to certain report, some chronic stress models cause the prefrontal cortex to release less glutamate (Son et al. 2018). The significant decrease in the glutamate and BDNF levels of the CUMS group observed in this study supports the aforementioned findings. This, however, runs counter to a publication (Li et al. 2018) that states that persistent unexpected stress is associated with impairments in synaptic proteins and buildup of extracellular glutamate. The disparity may be as a result of the differences in the CUMS models and a lot of other factors like the weight and sex of animal used. It is unclear exactly what modifications to glutamate transmission are brought on by stress, though. In this study, we examined how BDNF levels and glutamate release varied throughout CUMS and evaluated how these variations related to behaviors suggestive of depression. It was shown that only the CUMS rats showed signs of depression, such as decreased glutamate and BDNF levels. These results imply that vulnerability to stress-induced anhedonia may be correlated with aberrant release of glutamate and BDNF levels in the brain tissue. On the other hand, the GIRPOG-treated rats showed elevated levels of glutamate and BDNF, indicating this fraction's antidepressant properties. In a mouse model, 6-gingerol has been demonstrated to have the ability to combat depression (Sedighi et al. 2024).

There is connection between BDNF and oxidative stress, as oxidative stress has been reported to impair BDNF functions and causes psychiatric disorders (Shen et al. 2023). Oxidative stress indicators are elevated in response to prolonged stress (Che et al. 2015). The disruption of the anti-oxidative defense mechanism by CUMS has been documented.

Among the primary antioxidant molecules and enzymes that prevent oxidative stress are SOD and GSH, respectively. Our findings demonstrate that the brain tissues of the rats in CUMS had reduced levels of GSH and SOD activity, which is consistent with earlier observations. It is interesting to note that, in contrast to CUMS animals, 6-GIRPOG dramatically raised the SOD activity and GSH level in the brain tissues of the 6-GIRPOG-treated rats. Therefore, it may be said that 6-GIRPOG's antidepressant action against CUMS may be brought on by its anti-oxidative properties. This is consistent with a number of researches (Ghazizadeh et al. 2020; Akintoye et al. 2021; Asuku et al. 2023) that have documented the antioxidant properties of medicinal plants.

Oxidative stress has been associated with the triggering of pro-inflammatory signaling and cell death (Ajibare et al. 2023). Chronic stress exposure encourages persistent, non-resolving inflammation in the central nervous system, which could lead to depressive symptoms (Dionisie 2024). Elevated proinflammatory cytokine levels were seen in animal models of depression-like behavior; these were primarily TNF-α and IL-1β (Dai et al. 2020).

The HPA axis and sympathetic nervous system are triggered by social conflicts and stress. As seen in this study, the two work together to release norepinephrine, which raises the proinflammatory cytokines TNF-α, NFKB, IL-1β, and IL-6 (Dionisie 2024). Moreover, decrease in BDNF level has been reported to be responsible for the increase in production of brain pro-inflammatory markers (Ajibare et al. 2025). This corroborates the findings of the present study in CUMS-induced rats. However, study has reported that 6-GIRPOG has an anti-inflammatory effect,

which may account for the current study's observation of a decrease in pro-inflammatory cytokine levels following its administration (Ayinla and Asuku 2025). This could be because 6-GIRPOG has antidepressant properties that prevent the stimulation of the SNS and HPA axis, which in turn prevents the release of NE.

In conclusion, this study has been able to establish that CUMS-induced depressive like behavior in Wistar rats occurs through oxidative stress, inflammation and neurotransmission dysfunctions. Moreover, the anti-depressive effect of 6-GIRPOG on CUMS-induced rats is mediated by its anti-inflammatory, anti-oxidative, and neurotransmission modulatory abilities. In light of this, it is worthwhile to explore the great potential of 6-GIRPOG in providing panacea to the increasing incidence of depression. However, it has been reported that 6-GIRPOG had dose-dependent harmful impacts; hence, its use in pharmaceutical and food products should be controlled as it might be toxic at high doses (Chiaramonte et al. 2021). There is need for further investigations to examine the impact of 6-GIRPOG on mechanistic pathways in CUMS-induced rats, and its pharmacokinetic so as to translate these findings to humans.

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Conflicts of interest

The authors have declared that there is no conflict of interest.

Ethical Considerations

All the experiments complied with the ARRIVE Guidelines for preclinical animals studies and were approved by the Institutional Animal Care and Use Committee of the University of Ilorin, Nigeria (UERC/ASN/2023/2451).

Authors' Contributions

Research conception and design: MTA and AOA. Data collection: MTA and AOA. Analysis and interpretation of data: MTA and AOA. Drafting of the manuscript: AOA. Review and editing: MTA and AOA.

References

Ajibare AJ, Akintoye OA, Famurewa AC, Folawiyo MA, Bamisi OD, Asuku AO, Oyegbola OE, Christopher A, Olofinbiyi BA, Omotuyi OI (2023) Synergistic Action of Virgin Coconut Oil and Clomiphene in Reversing Endocrine Dysregulation in Letrozole-Model of Polycystic Ovarian Syndrome in Rats: Role of Nrf2/HMOX-1 Pathway. J Med Food, https://doi.org/10.1089/jmf.2023.0023.

Ajibare AJ, Akintoye OO, Oriowo OA, Asuku AO, Oriyomi IA, Adedara JF, Ayoola AM, Adejumo OM (2025) Dietary Virgin Coconut Exerted a Neuroprotective Effect via Upregulation of Nuclear Erythroid Factor and Downregulation Of Nuclear Factor KB Expression In Acrylamide-Induced Neurotoxicity: Role Of BDNF And AchE Pathways. TJNPR, 9(3): 1263 – 1269

Aryanezhad M, Abdi M, Amini S, Hassanzadeh K, Valadbeigi E, Rahimi K, Izadpanah E, Moloudi MR (2021) Cinnamomum zeylanicum extract has antidepressant-like effects by increasing brain-derived neurotrophic factor (BDNF) and its receptor in prefrontal cortex of rats. Avicenna J Phytomed, 11:302–313.

Asuku AO, Ayinla MT, Olajide TS, Yakub Z, Owoyele BV (2023) *Cola acuminata* Mitigates Cognitive Deficit and Oxidative Stress in Mercury Chloride-induced Neurotoxicity in Male Wistar Rats. Nigerian Journal of Neuroscience, 14(2):33-40.2023. http://doi.org/10.47081/njn2023.14.2/001

Asuku AO, Ayinla MT, Ajibare AJ, Olajide TS (2024) Mercury chloride causes cognitive impairment, oxidative stress and neuroinflammation in male Wistar rats: The potential protective effect of 6-gingerol-rich fraction of *Zingiber officinale* via regulation of antioxidant defense system and reversal of pro-inflammatory markers

increase. Brain Res. J, 148741: 1826, ISSN 0006-8993, https://doi.org/10.1016/j.brainres.2023.148741

Akintoye OO, Ajibare AJ, Fabunmi OA, Asuku AO, Tajudeen RA (2021) Effect of Honey on Oxidative Stress in Brain Tissue of Sleep Deprived Rats. Nig J Basic Appl Sci. 29(1): 83-89, DOI: http://dx.doi.org/10.4314/njbas.v29i1.10

Ayinla MT. Asuku AO (2025). The neurotoxic effects of lead acetate and the abrogating actions of 6-gingerol-rich extract of ginger via modulation of antioxidant defence system, pro-inflammatory markers, and apoptotic cascade. Naunyn-Schmiedeb. Arch. Pharmacol. 398, 9341-9345. https://doi.org/10.1007/s00210-025-03873-x

Bai Y, Song L, Dai G, Xu M, Zhu L, Zhang W, Jing W, Ju W (2018) Antidepressant effects of magnolol in a mouse model of depression induced by chronic corticosterone injection. Steroids, 135:73–78.

Benkermiche S, Djemli S, Haloui M, Benabed ML, Tahraoui A (2022) Preventive Effects of Ginger Extract and Nigella sativa Oil on Anxiety and Depression Behavior in Wistar Rats Exposed to Mercuric Chloride. Pharmacogn Res, 14(1):1-4.

Burstein O, Shoshan N, Doron R, Akirav I (2018) Cannabinoids prevent depressive-like symptoms and alterations in BDNF expression in a rat model of PTSD. Prog Neuropsychopharmacol Biol Psychiatry, 84:129–139.

Cavaleri D, Moretti F, Bartoccetti A, Mauro S, Crocamo C, Carrà G, Bartoli F (2023) The role of BDNF in major depressive disorder, related clinical features, and antidepressant treatment: insight from meta-analyses. Neurosci Biobehav Rev, 149:105159.

Che Y, Zhou Z, Shu Y, Zhai C, Zhu Y, Gong S, Cui Y, Wang, J-F (2015) Chronic unpredictable stress impairs endogenous antioxidant defense in rat brain. Neurosci Lett, 584, 208–213. https://doi.org/10.1016/j.neulet.2014.10.031

Chiaramonte M, Bonaventura R, Costa C, Zito F, Russo R (2021) [6]-Gingerol dose-dependent toxicity, its role against lipopolysaccharide insult in sea urchin (Paracentrotus lividus Lamarck), and antimicrobial activity. Food Biosci 39(100833):2212–4292.

Dai J, Pan J, Liao N, Shi J, Zeng Q, Huang L, Chen L (2020) Influence of miR-155 on behaviors of depression mice through regulating Wnt/β-catenin signaling pathway. Eur Rev Med Pharmacol Sci, 24, 1398–1407.

Dionisie V (2024) Inflammation, Oxidative Stress, and Chronic Unpredictable Mild Stress Model of Depression. JMHS, 2710-1452, 71-75, DOI: 10.32996/jmhs.

Gavzan H, Araghi A, Abbasabadi BM, Talebpour N, Golshahi H (2023) Antidepressant effects of a Persian herbal formula on mice with chronic unpredictable mild stress. Avicenna J Phytomed, 13(5): 562–574

Ghazizadeh J, Hamedeyazdan S, Torbati M, Farajdokht F, Fakhari A, Mahmoudi J, Araj-Khodaei M, Sadigh-Eteghad S (2020) *Melissa officinalis* L hydro-alcoholic extract inhibits anxiety and depression through prevention of central oxidative stress and apoptosis. Exp Physiol, 105:707–720.

Haq SH, AlAmro AA (2018) Neuroprotective effect of Quercetin in murine cortical brain tissue cultures, Clin Nutr Exp, 23, 89-96.

Keller J, Gomez R, Williams G, Lembke, A, Lazzeroni L, Murphy GM, Jr, Schatzberg AF (2017) HPA axis in major depression: cortisol, clinical symptomatology and genetic variation predict cognition. Mol Psychiatry, 22:527–536.

Kingir E, Sevinc C, Unal G (2023) Chronic oral ketamine prevents anhedonia and alters neuronal activation in the lateral habenula and nucleus accumbens in rats under chronic unpredictable mild stress. Neuropharmacology, 228, 109468.

Knezevic E, Nenic K, Milanovic V, Knezevic NN (2023) The Role of Cortisol in Chronic Stress, Neurodegenerative Diseases, and Psychological Disorders. Cells, 12, 2726. https://doi.org/10.3390/ cells12232726

Koch CEB, Leinweber BC, Drengberg C, Blaum, Oster H (2023) Interaction Between Circadian Rhythms and Stress. Neurobiol Stress 6: 57 – 67.

Kukula-Koch W, Koch W, Czernicka L, Glowniak K, Asakawa Y, Umeyama A (2018) MAO-A Inhibitory Potential of Terpene Constituents from Ginger Rhizomes—A Bioactivity Guided

Fractionation. Molecules, 23(6): 1301. https://doi.org/10.3390/molecules23061301.

Lee DY, Kim E, Choi MH (2015) Technical and clinical aspects of cortisol as a biochemical marker of chronic stress. BMB Rep. 2015, 48, 209–216.

Li SX, Han Y, Xu LZ, Yuan K, Zhang RX, Sun CY, Xu DF, Yuan M, Deng JH, Meng SQ, Gao XJ, Wen Q, Liu LJ, Zhu WL, Xue YX, Zhao M, Shi J, Lu L (2018) Uncoupling DAPK1 from NMDA receptor GluN2B subunit exerts rapid antidepressant-like effects. Mol Psychiatry, 23:597–608.

López-López AL, Herlinda BJ, María DEV, Mailinalli BP, Gonzalo VP, dan Fransisco JAA (2016) Chronic Unpredictable Mild stress Generates Oxidative Stress and Systemic Inflammation in Rats. Physiol Behav, 161: 15-23.

Murrough JW, Abdallah CG, Mathew SJ (2017) Targeting glutamate signalling in depression: progress and prospects. Nat Rev Drug Discov, 16:472–486.

Phukan S, Adhikari K (2017) Study of the antidepressant and antinociceptive activity of ethanolic extract of rhizomes of *Zingiber officinale* in experimental animals. Int J Pharm Sci Res, doi:10.13040/IJPSR.0975-8232.8(7).3004-09

Rafeyan R, Papakostas GI, Jackson WC, Trivedi MH (2020) Inadequate response to treatment in major depressive disorder: Augmentation and adjunctive strategies. J Clin Psychiatry, 81:OT19037BR3.

Sedighi S, Nasiri B, Alipoor R, MoradiKor N (2024) Modulation of 6-Gingerolin Antidepressant-like Effects in Mice Model. GMJ Medicine, 3(2):59-63.

Shalinee S, Meraj K, Ritika S (2022) Evaluation of Antidepressant Effect of 6-Shogaol in animal models, IJNRD, 7 (12): 2456-4184.

Shen R, Ardianto C, Celia C, Sidharta, VM, Sasmita PK, Satriotomo I, Turana Y (2023) Brain-derived neurotrophic factor interplay with oxidative stress: neuropathology approach in potential biomarker of Alzheimer's disease. Dement Neuropsychol. 4:17, 20230012.

Son H, Baek JH, Go BS, Jung DK, Sontakke SB, Chung HJ, Lee DH, Roh GS, Kang SS, Cho GJ, Choi WS, Lee DK, Kim HJ (2018) Glutamine has antidepressive effects through increments of glutamate and glutamine levels and glutamatergic activity in the medial prefrontal cortex. Neuropharmacology, 143:143–152.

Vaváková M, Ďuračková Z, Trebatická J (2015) Markers of oxidative stress and neuroprogression in depression disorder. Oxid Med Cell Longev. 2015:898393

Wei H, Zhou T, Tan B, Zhang L, Li M, Xiao Z, Xu F (2017) Impact of chronic unpredicted mild stress-induced depression on repaglinide fate via glucocorticoid signaling pathway. Oncotarget, 8:44351–44365.

World Health Organisation (2017) Depression and Other Common Mental Disorders Global Health Estimates. World Health Organization; Licence: CC BY-NC-SA 3.0 IGO.

World Health Organisation (2021) Depression. Fact sheet. Geneva: WHO. Available at: https://www.who.int/news-room/fact-sheets/ detail/depression.