Current Pharmaceutical Research (CPR)
Vol 1, Issue 4, October-December 2025
Journal Homepage: https://cpr.org.in/

Research Article

Neurobehavioral and Antioxidant Activity of Crassula ovata Extract in Haloperidol-Induced Alterations in Albino Rats

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Keywords

Crassula ovata; Haloperidol; Neurobehavioral activity; Oxidative stress; Superoxide dismutase; Albino rats.

Abstract

Haloperidol, a typical antipsychotic drug, is widely used in the treatment of schizophrenia but is associated with neurobehavioral impairments and oxidative stress upon prolonged administration. The present study aimed to evaluate the neurobehavioral and antioxidant potential of the ethanolic extract of Crassula ovata in haloperidol-induced alterations in albino rats. Animals were subjected to haloperidol-induced neurotoxicity and treated with C. ovata extract at doses of 200 and 400 mg/kg, with imipramine serving as the standard reference drug. Neurobehavioral assessments were performed using the open field test and cage climbing test to evaluate locomotor activity, exploratory behavior, and motor coordination. Oxidative stress was assessed by estimating superoxide dismutase (SOD) activity in brain tissue. Haloperidol administration significantly reduced locomotor activity, exploratory behavior, climbing duration, and endogenous antioxidant defenses. Treatment with C. ovata extract produced a dose-dependent improvement in behavioral parameters and significantly elevated SOD activity compared with the disease control group. The higher dose (400 mg/kg) demonstrated effects comparable to the standard drug. These findings suggest that Crassula ovata possesses significant neuroprotective and antioxidant properties, supporting its potential therapeutic role in managing drug-induced neurobehavioral and oxidative impairments.

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Article Info
Received: 08 December 2025; Received in revised form: 16 January 2026; Accepted: 16 January 2026; Available online: 21 January 2026; Volume: 1; Issue: 4; Pages: 491-497.

ISSN: 3049-2955/The authors © 2025, under exclusive license to the Sprout Publication.
DOI: https://doi.org/10.63785/2025.1.4.491497

1. Introduction

Oxidative stress plays a pivotal role in the pathogenesis of several neuropsychiatric and neurodegenerative disorders. An imbalance between the production of reactive oxygen species and endogenous antioxidant defense mechanisms leads to neuronal damage, altered neurotransmission, and behavioral impairments [1]. Increasing evidence suggests that antipsychotic drugs, particularly with prolonged use, can exacerbate oxidative stress in the central nervous system, thereby contributing to motor dysfunction and cognitive deficits [2].

Haloperidol, a widely prescribed typical antipsychotic, is commonly used in the management of schizophrenia and other psychotic disorders. However, chronic administration of haloperidol is associated with extrapyramidal side effects, reduced locomotor activity, and neurobehavioral alterations [3]. These adverse effects are largely attributed to dopamine receptor blockade and enhanced oxidative burden in brain tissue. Consequently, haloperidol-induced models are extensively employed to evaluate neuroprotective, behavioral, and antioxidant interventions in experimental animals [4].

In recent years, growing interest has been directed toward natural products as potential neuroprotective agents due to their safety profile and diverse pharmacological activities. Medicinal plants rich in secondary metabolites such as flavonoids, alkaloids, steroids, and phenolic compounds have demonstrated antioxidant and neuromodulatory properties [5]. *Crassula ovata*, a succulent plant belonging to the family Crassulaceae, is traditionally used for various therapeutic purposes. Phytochemical investigations have reported the presence of bioactive constituents

that may confer antioxidant and cytoprotective effects [6].

Although *C. ovata* has shown antioxidant activity in vitro, limited data are available regarding its in vivo neurobehavioral and antioxidant potential [7]. Therefore, the present study was designed to evaluate the protective effects of *Crassula ovata* extract against haloperidol-induced behavioral impairments and oxidative stress in albino rats, using behavioral assessments and biochemical estimation of superoxide dismutase activity [8].

2. Materials and Methods

2.1 Plant Material

Fresh leaves of *Crassula ovata* were collected and authenticated at the ABS Botanical Garden, Salem, Tamil Nadu, India (Reference No. AUT/SSMC/304). The collected plant material was thoroughly washed, shade dried at room temperature, and coarsely powdered using a mechanical grinder. The powdered leaves were stored in airtight containers under appropriate conditions until further extraction.

2.2 Preparation of Extract

The powdered leaf material of *Crassula ovata* was subjected to Soxhlet extraction using ethanol as the solvent. The extraction process was carried out until complete exhaustion of the plant material. The resulting extract was filtered and concentrated under reduced pressure to obtain a dry residue. The dried extract was then stored in an airtight container under appropriate conditions until further analysis [9], [10], [11].

2.3 Experimental Animals

Healthy albino rats of either sex, weighing between 150 and 200 g, were used for the experimental study. The animals were housed under standard laboratory conditions with controlled temperature, humidity, and a 12 h light–dark cycle, and were provided free access to standard pellet diet and water ad libitum. All experimental procedures were conducted in accordance with the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals [12], [13].

The experimental protocol was reviewed and approved by the Institutional Animal Ethics Committee. Ethical approval was obtained under IAEC Registration No. 2255/PO/Re/S/23/CPCSEA, with Proposal No. SSMCOP/IAEC/M.Pharm/05/06/2025.

2.4 Experimental Design

The animals were randomly divided into five experimental groups, with six rats in each group. Group I served as the normal control, while Group II received haloperidol (10 mg/kg, i.p.) as the disease control. Group III was treated with haloperidol along with the standard drug imipramine (25 mg/kg). Groups IV and V received haloperidol in combination with the ethanolic extract of *Crassula ovata* at doses of 200 mg/kg and 400 mg/kg, respectively. Behavioral and biochemical parameters were assessed during the experimental period [14], [15], [16].

Table 1 presents the grouping pattern, treatments administered, and corresponding doses used in the study to evaluate the protective effect of *Crassula ovata* against haloperidol-induced behavioral alterations [17], [18].

Table 1: Experimental design and treatment protocol.

Group	Treatment	Dose & Route
Group I	Normal control	—
Group II	Haloperidol control	10 mg/kg, i.p.
Group III	Haloperidol + Imipramine	25 mg/kg
Group IV	Haloperidol + *C. ovata*	200 mg/kg
Group V	Haloperidol + *C. ovata*	400 mg/kg

(n=6)

2.5 Behavioral Assessments

2.5.1 Open Field Test

The open field test was used to assess locomotor and exploratory behavior in rats. Animals were placed individually in an open field arena, and the time spent in the central area and total distance travelled were recorded. Increased central zone activity was considered indicative of reduced anxiety and enhanced exploration [19]. Haloperidol-treated rats showed a significant reduction in exploratory behavior compared to normal controls. Treatment with imipramine markedly improved locomotor activity. Administration of the ethanolic extract of *Crassula ovata* at doses of 200 mg/kg and 400 mg/kg produced a dose dependent increase in central zone exploration, with the higher dose exhibiting effects comparable to the standard drug [20].

Table 2 summarizes the effect of different treatments on central zone activity in the open field test, reflecting changes in exploratory behavior and anxiety levels among experimental groups [21].

Table 2: Effect of *Crassula ovata* on time spent in the central zone in the open field test.

Group	Time spent in center (seconds)
Group I	100–120
Group II	75–95
Group III	170–185
Group IV	150–170
Group V	180–210

2.5.2 Cage Climbing Test (CCT)

The cage climbing test was performed to assess motor activity and extrapyramidal effects in experimental rats [22]. Each animal was placed individually in a wire mesh cage, and the total duration of climbing behavior was recorded over a period of five minutes. A reduction in climbing activity was considered indicative of haloperidol-induced motor suppression and extrapyramidal dysfunction [23].

2.6 Biochemical Estimation

Superoxide dismutase activity in brain tissue was quantified to assess oxidative stress status in experimental animals [24]. The assay was performed using standard spectrophotometric methods, and the optical density of the sample and control was measured [25]. The percentage of SOD activity was calculated using the following equation:

$$\text{SOD activity (\%)} = \left[1 - \left(\frac{OD_{sample}}{OD_{control}}\right)\right] \times 100$$

The obtained values were used to evaluate the antioxidant potential of the treatments and their ability to mitigate haloperidol-induced oxidative stress [26].

3. Results

3.1 Open Field Test & Time Spent in Center (Seconds)

The effect of *Crassula ovata* on locomotor and exploratory behavior was evaluated using the open field test. Time spent in the central zone was recorded as an indicator of anxiety-like behavior and exploratory activity. The results are presented in Figure 1.

Figure 1 Effect of *Crassula ovata* on time spent in the central zone during the open field test. The figure depicts the mean time spent in the central area by animals from different experimental groups. The haloperidol-treated group shows a marked reduction in central zone activity compared with the normal control, indicating suppressed locomotor and exploratory behavior. Treatment with the standard drug imipramine significantly increased the time spent in the center. Animals treated with the ethanolic extract of *Crassula ovata* at doses of 200 mg/kg and 400 mg/kg exhibited a dose dependent increase in central zone exploration, with the higher dose showing the greatest improvement, comparable to or exceeding the standard treatment. Error bars represent variability within each group.

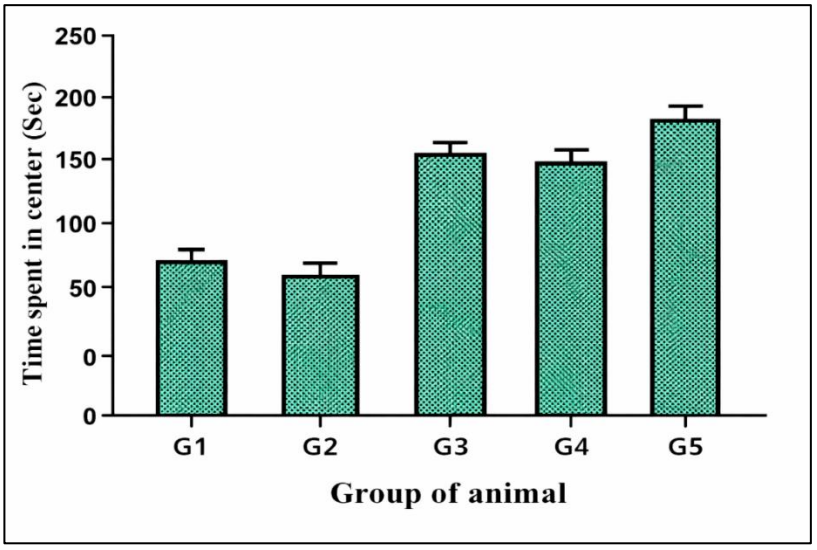

Figure 1: Time spent by test animals in Open Field Test.

Interpretation

Haloperidol treatment resulted in a reduction in total distance travelled, indicating suppressed locomotor activity. Administration of the ethanolic extract of *Crassula ovata* at both tested doses significantly improved locomotor performance, with the 400 mg/kg dose showing the greatest enhancement. The observed increase in distance travelled suggests a reversal of haloperidol-induced motor suppression.

Table 3 summarizes the total distance travelled by animals in different experimental groups during the open field test. The haloperidol-treated group exhibited reduced locomotor activity compared with the normal control. Treatment with imipramine and *C. ovata* resulted in increased distance travelled, with the higher dose of *C. ovata* demonstrating maximal improvement.

Table 3: Effect of *Crassula ovata* on total distance travelled in the open field test.

Group	Range (seconds)
Group I	770–800 sec
Group II	700–730 sec
Group III	750–800 sec
Group IV	750–800 sec
Group V	790–850 sec

Figure 2 illustrates the effect of various treatments on total distance travelled in the open field test. Haloperidol significantly reduced locomotor activity, whereas imipramine and Crassula ovata treatment increased movement in a dose dependent manner. The higher dose of C. ovata produced locomotor activity comparable to the standard drug, indicating its protective effect against haloperidol-induced motor impairment.

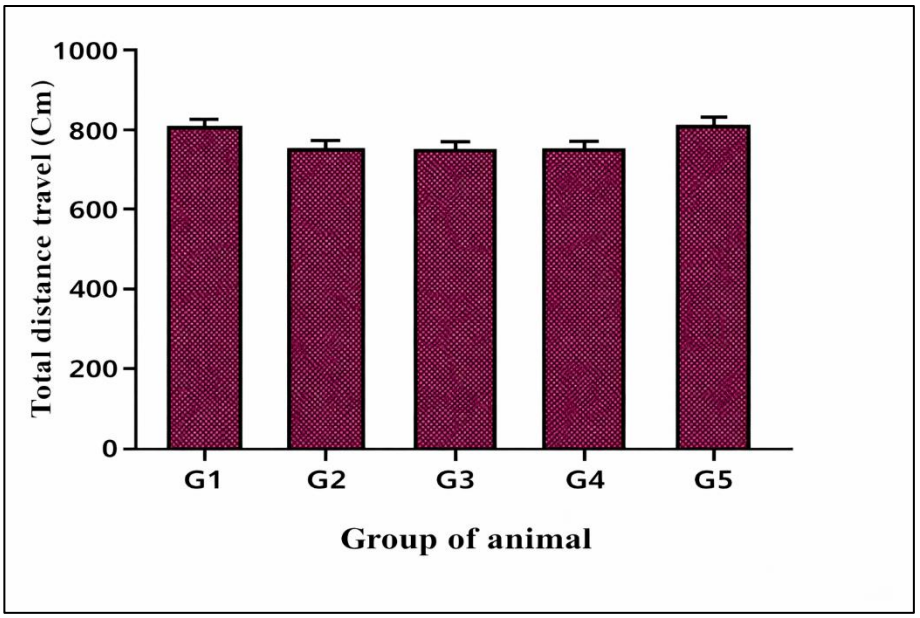

Figure 2: Distance travelled by test animals in the open field test.

Interpretation
Reduced locomotor activity in the haloperidol control group indicates motor suppression. Treatment with *Crassula ovata* extract increased locomotion in a dose-dependent manner, suggesting a protective effect against haloperidol-induced motor impairment.

3.2 Cage Climbing Test
Haloperidol treatment markedly reduced climbing duration, indicating significant motor suppression. Rats treated with the standard drug imipramine and the ethanolic extract of Crassula ovata exhibited a noticeable improvement in climbing activity. Group III and Group V showed the highest climbing times, while Group IV demonstrated a moderate but significant increase compared with the haloperidol control group. These findings suggest that C. ovata effectively attenuates haloperidol-induced motor deficits.

3.3 Superoxide Dismutase Activity
Haloperidol administration significantly reduced endogenous antioxidant defense, as evidenced by decreased SOD activity in the disease control group. Treatment with the standard drug imipramine markedly restored SOD levels. Rats treated with the ethanolic extract of *Crassula ovata* showed a dose dependent increase in SOD activity, with the 400 mg/kg dose producing a greater antioxidant response than the 200 mg/kg dose. These findings indicate the ability of *C. ovata* to attenuate haloperidol-induced oxidative stress.

Table 4 presents the percentage SOD activity observed in different experimental groups. The haloperidol-treated group showed reduced antioxidant activity, while treatment with imipramine and *C. ovata* significantly elevated SOD levels, particularly at the higher dose.

Table 4: Effect of *Crassula ovata* on SOD activity in brain tissue.

Group	Percentage (%)
Group I	32.78%
Group II	22.78%
Group III	45.34%

Group IV	37.88%
Group V	40.29%

Figure 3 illustrates the optical density values measured at 420 nm during SOD estimation. Increased absorbance in extract-treated groups reflects enhanced SOD activity compared with the haloperidol control group, demonstrating the antioxidant potential of *Crassula ovata*.

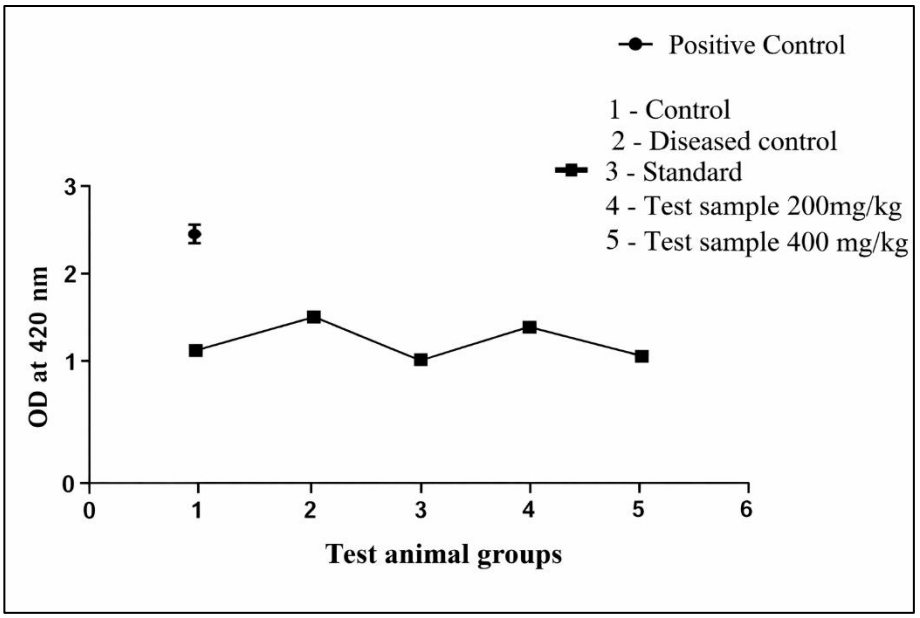

Figure 3: Optical density at 420 nm during SOD activity estimation.

Interpretation
Haloperidol suppressed endogenous antioxidant defenses, whereas treatment with *Crassula ovata* significantly elevated SOD activity in a dose dependent manner, indicating its protective antioxidant effect.

4. Discussion
Haloperidol administration is known to induce motor impairment and oxidative stress, which was evident in the present study through reduced open field test performance, diminished cage climbing behavior, and decreased superoxide dismutase activity. These findings confirm the successful induction of neurobehavioral deficits and oxidative imbalance. Treatment with the ethanolic extract of *Crassula ovata* significantly reversed these alterations, indicating its protective potential [27].

The improvement observed in open field test parameters suggests enhanced locomotor and exploratory behavior along with reduced anxiety-like responses in extract-treated animals. Increased climbing duration further indicates a protective effect on dopaminergic pathways, which are typically suppressed by haloperidol-induced dopamine receptor blockade. Restoration of climbing behavior implies attenuation of extrapyramidal motor deficits [28].

Additionally, the marked elevation in SOD activity following *C. ovata* treatment reflects its antioxidant capability. Enhanced antioxidant defense may contribute to the reduction of oxidative stress and restoration of neurochemical balance, thereby improving motor and behavioral outcomes. Among the tested doses, the 400 mg/kg extract produced effects comparable to the standard drug, highlighting its strong neuroprotective and antioxidant potential [29].

These findings collectively suggest that *Crassula ovata* may offer therapeutic benefits against drug-induced neurotoxicity, warranting further investigations to elucidate its active constituents and underlying mechanisms of action [30].

Conclusion
The ethanolic extract of *Crassula ovata* produced significant improvements in behavioral performance and antioxidant status in haloperidol-treated rats. Restoration of locomotor activity, climbing behavior, and superoxide dismutase levels indicates its neuroprotective and antioxidant efficacy. Among the tested doses, 400 mg/kg exhibited the most pronounced effects, comparable to the standard drug. These findings support the potential application of *C. ovata* as a natural neuroprotective and antioxidant agent, warranting further studies to identify its active constituents and elucidate underlying mechanisms.

Acknowledgment
The authors sincerely acknowledge their respective institutions for providing the necessary facilities and academic support to carry out this review work. The authors are also grateful to colleagues and peers for their constructive suggestions during the preparation of the manuscript.

Author Contribution
SN; Conceptualization of the review, **SK**; Manuscript drafting, **BS**; Supervision and final approval.

Source of Funding
None

Ethical Approval
All animal experiments were conducted in accordance with CPCSEA guidelines and were approved by the Institutional Animal Ethics Committee (IAEC Reg. No. 2255/PO/Re/S/23/CPCSEA; Proposal No. SSMCOP/IAEC/M.Pharm/05/06/2025).

Data Availability
The data generated and analyzed during this study are available from the corresponding author upon reasonable request.

Conflicts of Interest
There are no conflicts of interest

Declarations
The authors declare that they used AI language tools (ChatGPT and Grammarly Premium) to enhance this manuscript's linguistic clarity and readability. They carefully reviewed and edited all generated text to ensure accuracy and alignment with the research's intended meaning.

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