Contents lists available at UGC-CARE

International Journal of Pharmaceutical Sciences and Drug Research

[ISSN: 0975-248X; CODEN (USA): IJPSPP]

journal home page : http://ijpsdronline.com/index.php/journal

Research Article

Comparative Study of Neuroprotective Activity of *Mucuna pruriens* and *Withania somnifera* Against Haloperidol-Induced Parkinson's Disease in an Animal Model

Mary Brenda Ogwang, Deepak Kumar Jha*, Sandipan Chatterjee

Department of Pharmacology, Karnataka College of Pharmacy, Bengaluru, India.

ARTICLE INFO

Article history:

Received: 28 July, 2025

Revised: 29 August, 2025

Accepted: 07 September, 2025

Published: 30 September, 2025

Keywords:

Biochemical, Behavioural assay, *Mucuna pruriens* seeds, MTT assay, Neuroprotective, *Withania somnifera*

DOI:

10.25004/IJPSDR.2025.170505

ABSTRACT

Parkinson's disease (PD) initially was a challenging neurodegenerative disorder (ND) to treat and manage, but translational research bridged the gap to its risk factors, pathophysiology, diagnosis, treatment and management. Research is more focused on neuroprotection with the purpose of preventing neuronal loss reaching more than 60% exhibiting symptomatic PD, and neuro-rescue to restore functionality to partly damaged cells. Thus, this comparative study investigated *Mucuna pruriens* and *Withania somnifera's* neuroprotective properties, as this may help to uncover and establish similar pathways in their mechanisms of action, and/or differences to determine if synergism may be possible. The administration of haloperidol resulted in significant motor damage, oxidative stress, and high pro-inflammatory cytokines. *M. pruriens* and *W. somnifera* extract pre-treatment showed dose-dependent beneficial effects on behavior parameters, enhancing GSH levels and decreasing LPO levels, and remarkably reduced the levels of inflammatory cytokines. These results were corroborated by histopathological analysis of brain tissues, which indicated well-preserved brain neuronal integrity and limited evidence of neurodegeneration. Moreover, the *in-vitro* MTT test using neuro-2a cells showed that both extracts were able to support high cell viability; it is implied that the extracts may possess cytoprotective potentials on the cellular level. The experiment shows that *M. pruriens* and *W. somnifera* have substantial neuroprotective and anti-inflammatory effects on haloperidol-induced neurotoxicity, *W. somnifera* having a better overall outcome as compared to *M. pruriens*.

Introduction

Parkinson's disease (PD) is a chronic neurological condition that causes an impairment of movement, mental well-being, sleep, and other physical processes. It is predominant in older people, though there might be a tendency for occurrence among the youth, with more men than women being affected. However, although it is incurable, symptoms that include tremor, rigidity, slow movement, and contractions can be suppressed using medications or even therapy, including levodopa/carbidopa, physiotherapy, and deep brain stimulation. Other non-motor symptoms caused by PD include thinking difficulties, memory loss, depression, sleep disturbances and many people have high disability rates and tend to need care. The world is facing the rising burden of PD, whose prevalence has doubled in the last 25 years especially in economically lower- and middle-tier countries where treatment, medicine, and diagnosis are still not accessible. The WHO highlights the value of reinforcing health systems, ensuring greater access to treatment, assisting caregivers, and countering stigma and misconceptions about the illness.[1] PD begins with a preclinical phase exhibiting non-motor symptoms, including olfactory dysfunction, depressive and anxiety symptoms, and disrupted sleep, and autonomic dysfunction, often lasting years before motor symptoms appear. The clinical phase begins with motor signs like resting tremor, bradykinesia, rigidity, and balance problems, typically progressing

***Corresponding Author:** Dr. Deepak Kumar Jha

Address: Department of Pharmacology, Karnataka College of Pharmacy, Bengaluru, India.

Email ✉**:** deepakjha736@gmail.com

Tel.: +91-7411458812

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2025 Mary Brenda Ogwang *et al*. This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

gradually and asymmetrically. Current therapies, including levodopa, offer symptom relief but lose efficacy over time and may cause complications like dyskinesia, dystonia, and cognitive decline. A major challenge is the late diagnosis, as significant neurodegeneration occurs before clinical signs appear. Therefore, developing early diagnostic biomarkers and new therapeutic strategies remains a critical research priority to delay or prevent progression.[2] PD is a complicated disorder and differs substantially among individuals, so individualized care is paramount. The priorities of NINDS-funded research are on early detection, disease sub-phenotyping and development of neuroprotective interventions, including calcium channel blockers and upregulation of parkin protein expression. GDNF-based gene therapy, stem-cell-based dopamine replacement and immune-modulating drugs such as sargramostim are also in development. Deep brain stimulation is one of the most relevant therapies, and attempts to make it more accurate and investigate some alternatives to it, such as non-invasive tDCS, continue. Imaging, cueing, and a behavioral intervention are some ways that researchers are dealing with drug-resistant symptoms, including dyskinesia, dementia, sleep disorders, and freezing of gait. Adaptive technologies, mobile health tools, and exercises are being created to help better control the symptoms and quality of life.[3]

Plant Information

Herbal plants are used in the management of Parkinson's disease based on the fact that they provide neuroprotective, anti-inflammatory, antioxidant and dopaminergic effects, which are all applicable in the pathogenesis and management of the symptoms of PD. Oxidative stress is a major contributing factor in dopaminergic degeneration produced by PD. The antioxidants found in many herbs are polyphenols, flavonoids, and other free-radical scavengers (e.g., *Curcuma longa* [turmeric], *Camellia sinensis* [green tea]).[4,5] PD progressing with chronic neuroinflammation is known. The *Withania somnifera* (Ashwagandha), *C. longa* are herbs and are known to have anti-inflammatory activities through the regulation of cytokines and blocking NF- kβ chains. Some plant extracts promote the action of mitochondria and block apoptosis in neurons. *Bacopa monnieri* and *Ginkgo biloba* have been effective in improving memory, cognitive efficiency and neuronal degeneration.[6-9] There are herbs that can either raise the dopamine levels or have dopaminergic actions. Instead of synthetic levodopa, some more natural sources of L-DOPA, such as *Mucuna pruriens*, which is a direct precursor of dopamine itself, have proven equally or even more tolerable than conventional levodopa in research. *M. pruriens,* commonly referred to as cowage, velvet bean, or Alkushi, is a tropical legume native to Africa and tropical Asia, and it is widely cultivated and naturalized in many regions.[10,11] Herbal remedies can be used as safer and tolerable substitutes or adjuncts to the standard PD drugs. They are appropriate considering their multitarget activities, which can address motor as well as non-motor manifestations, including anxiety, sleep disturbances, and fatigue. There are numerous botanical preparations that have been shown to be neuroprotective in animal models of PD There are some clinical results of quality-of-life, motor-function, and cognitive symptoms improvement because of applying herbal formulations in adjunct therapies. Herbal plants can potentially provide a multimodal management of PD given their potential activities on major pathogenic processes, including oxidative stress, inflammation, mitochondrial aberrations, and dopamine depletion. Despite the potential, these therapies are still more standardized, clinically tested and regulated in order to gain popularity in evidence-based medicine. Parkinson's disease is experienced in over 1.0% of people aged 65 and over[12] and Alzheimer's Disease is experienced in up to 10.0% of the same people.[13] This necessitates a re-evaluation of existing therapeutic agents currently used for symptomatic management of neurodegenerative conditions such as PD, AD, Huntington's disease, and ALS[14] and focuses on the research that is involved in the determination of therapeutic agents that alter the prognosis of the neurodegenerative diseases by inhibiting the death of neurons or by promoting the repair of neurons.

Research Hypothesis

Experimental models of PD play a crucial role in evaluating potential neuroprotective agents. Haloperidol, a typical antipsychotic, is widely used to induce Parkinsonism in animals by producing dopaminergic blockade and oxidative stress, thereby mimicking gradual disease progression. This model allows the assessment of neuroprotective interventions targeting oxidative stress, mitochondrial dysfunction, and neuroinflammatory pathways. Herbal medicines are gaining increasing attention as sources of neuroprotective agents. *M. pruriens* and *W. somnifera,* important adaptogenic plants in Ayurvedic practice, have demonstrated neuroprotective potential through their antioxidant, anti-apoptotic, and immunomodulatory properties. However, direct comparative evaluation of their efficacy against haloperidol-induced neurotoxicity has not been adequately explored. Hence, this study aimed to evaluate and compare the neuroprotective efficacy of ethanolic extracts of *M. pruriens and W. somnifera* against haloperidol-induced Parkinsonism. The investigation involved phytochemical screening, neurological behavioral testing, evaluation of biochemicals, assessment of inflammatory mediators, and mitochondrial activity, thereby providing a comprehensive insight into their neuroprotective and immunomodulatory properties. The results of this study are anticipated to enhance understanding of natural neuroprotective agents.

Materials And Methods

Collection of Materials

The plant materials were collected from an authenticated vendor (Chisthi Mangalore Ayurveda) and verified by Dr. Noorunnisa Begum, Curator, FRLHT, Bengaluru, India. The authentication was documented under registration numbers 5891 and 6755.

Preparation of Extract

The plant parts (*M. pruriens*, seeds and *W. somnifera, leaves*) were dried under shade a room temperature. Drying of the materials proceeded then by powdering them into a coarse sieve (10/40 mesh). About 100 g of this powdered material was then placed under ethanolic extraction. About 1000 mL of ethanol was brought to be extracted in a reflux condenser, which was performed three times for 7 hours or until the reduction to half of the volume. Whatman filter paper No. 1 was used to filter the extract and left to evaporate to give the constant weight.[15] The dry residue was further subjected to an initial phytochemical screening to confirm the presence of bioactive constituents.

Experimental Design

The study used male *Wistar* rats of integrated weight and they weighed about 150 to 200 g. The animals were purchased according to the approved Animal Research Facility and stored in polypropylene cages with 6 to 8 rats kept in each cage. In preparation for the experiment, the rats were acclimatized to the laboratory environment over a one-week period with normal conditions in terms of light, a range of 12 hours in the light and 12 hours in darkness. They were provided with a normal rodent diet, which was bought at a commercial store and supplied with water ad libitum. Any action on animals followed the regulations of the CCSEA and was accepted by the IAEC (KCP/IAEC/11/22-23/04/22/12/22).

The study was conducted using seven groups of Wistar rats (n=6 per group). Group I served as the normal control and received Saline (2 mL/kg/day, p.o.) for 21 days. Group II (disease control) received haloperidol (2 mg/kg/day, p.o.).[16] Group III (standard) received levodopa (15 mg/kg/day, p.o.)[17] followed by haloperidol one hour later. Groups IV and V were given *M. pruriens* extract at 200 and 400 mg/kg, respectively, one hour prior to haloperidol. Groups VI and VII received *W. somnifera* extract at 100 and 200 mg/kg, respectively, also one hour before haloperidol. All treatments were administered orally for 21 consecutive days.

Dose Selection

The doses of *M. pruriens* and *W. somnifera* extracts were selected based on previously reported toxicity studies in rats [Krishna *et al*., 2016; Balkrishna *et al.,* 2022].

Neurological, Behavioral and Biochemical Analysis

Motor coordination rotarod test in accordance with *Manna et al.* (2006),[18] The single-pole evaluation of bradykinesia according to *Hu et al.* (2018),[19] hanging test (according to *Rai et al.,* 2016)[20] in which rats were placed on a horizontal grid, which was inverted to determine grip endurance.

Animals were sacrificed 48 hours after the final administration of the drug at the end of the experimental period. Pentobarbitone in a high dose was used to facilitate euthanasia, and brains were dissected and analyzed biochemically and histopathological. Quantitation of glutathione (GSH) by colorimetric measurement (Ellman 1959; *Jollow et al.* 1974)[21] of its content as DTNB. Evaluation of lipid peroxidation (LPO) was determined based on the procedure developed by Ohkawa *et al.* (1978).[22] ELISA kits measured the levels of cytokines (IL-1beta, TNF-alpha, and IL-6) following the guidelines of Farombi *et al*. (2019) and Mir *et al.* (2025).[23,24]

Histopathological Analysis of the Brain

The brains of the rats were gently extracted and held in a petri dish after the experiment was done. The brain was rapidly dissected to minimize tissue degradation by chopping away the hippocampus and the frontal cortex. The brain parts isolated immediately after the dissection process were placed in 10% formalin to fix the tissue, ready to be examined histologically.[25]

Methodology for Cell Viability Calculation (MTT Assay)

The neuro-2α neuroblastoma cell line has been utilized in the study of the toxicity of cell lines. The cells used were purchased at NCCS, Pune and the passage no. N2a-P176. The cells were grown in DMEM. The survival of cells was determined by the MTT assay, which indicated a percentage of survival around 84 to 85% at P1, which is a measure of mitochondrial metabolic activity. Cell suspension (2×10^3 cells/well) was plated in 96-well plates and cultured for 24 hours. The cells were further incubated at different concentrations of different formulations at three replicates each after 24 hours of incubation. After treatment, MTT reagent (20 µL) was inserted into each well. Active-cell mitochondrial dehydrogenases metabolize UV-sensitive dye-based resazurin, MTT, or resazurin to insoluble purple crystals of formazan. The medium was removed after a period of 4 hours of incubation and 100 µL of DMSO was added to dissolve the formazan in each well. The plate was gently shaken at room temperature (5 minutes). A spectrophotometer was used to measure absorbance at 545 nm.[26]

The viability (%) of cells was determined with the following formula:

Cell Viability = (Absorbance of treated cells/Absorbance of untreated control cells) X 100

Statistical Analysis

The data are observed as Mean ± S.E.M., n=6. We conducted the statistical analysis using GraphPad. In the case of statistics, ANOVA and Tukey were applied to make multiple comparisons within the groups. The value of *p* less than 0.05 was considered statistically significant. These findings suggest that the treatments had a notable impact on the measured outcomes, warranting further investigation into their underlying mechanisms. 0.05 was considered statistically significant, comparing the untreated normal control group with the others.

Results

Phytochemical Analysis

M. pruriens is rich in phytochemicals, notably tannins, saponins, phenolic compounds, and various proteins and carbohydrates. *W. somnifera* is rich in steroids and alkaloids, along with flavonoids, tannins, and phenolic compounds.

Neurological Behavioral Assay

Fig. 1 shows the rotarod performance over 21 days for all treatment groups. Haloperidol latency induces motor dysfunction. Standard drugs, L-DOPA and *M. pruriens,* show significant recovery, suggesting dopaminergic restoration. *W. somnifera* shows weak to moderate effects, which may not be sufficient alone for motor recovery at tested doses but can be added to the main course of the treatment. Fig. 2 shows the pole test results over time (Day 0–21). Haloperidol impaired motor function initially, and the effect was reversed by the standard drug L-DOPA, but it showed increased time at later stages. The test drugs, *M. pruriens* and *W. somnifera,* demonstrated neuroprotective or dopaminergic potential, with more stable and lower descent times than L-DOPA in later phases. *Mucuna* 400 mg/kg showed the most consistent improvement, suggesting a dose-dependent benefit. Fig. 3 shows the HANGING Test (inverted 16 cm above a flat surface). Haloperidol induces motor impairment, reflected by very low hanging times. L-DOPA and the test drugs *M. pruriens* and *W. somnifera* improve motor function significantly. Both herbal treatments show dose-dependent improvements, with higher doses performing closer to normal controls. Fig. 4 shows the percentage of success in neurobehavioral tests (hanging, pole, and rotarod) for each treatment group. Haloperidol significantly impaired motor function across all tests. *W. somnifera* (200 mg/kg) and *M. pruriens* (400 mg/kg) showed strong potential in improving neuromuscular performance, especially in the hanging test. The pole test reflects a more resistant motor deficit (bradykinesia), with minimal recovery across treatments. Rotarod improvements were modest, but *Mucuna* 400 mg/kg had the best outcome among treatments.

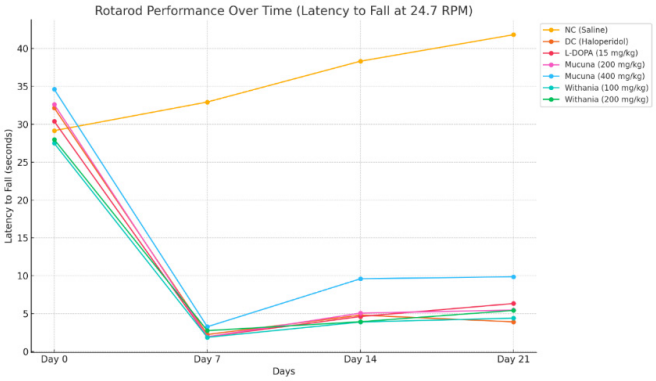


Fig. 1: Rotarod Performance over 21 days for all treatment groups. Values are expresses as a Mean ± S.E.M/ n = 6

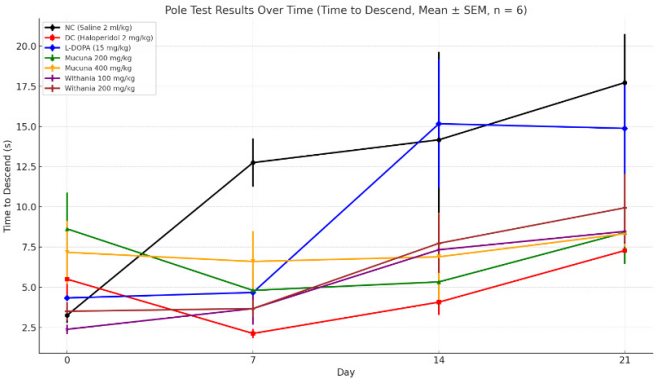


Fig. 2: Pole Test over 21 days for all treatment groups. Values are expresses as a Mean ± S.E.M/ n = 6

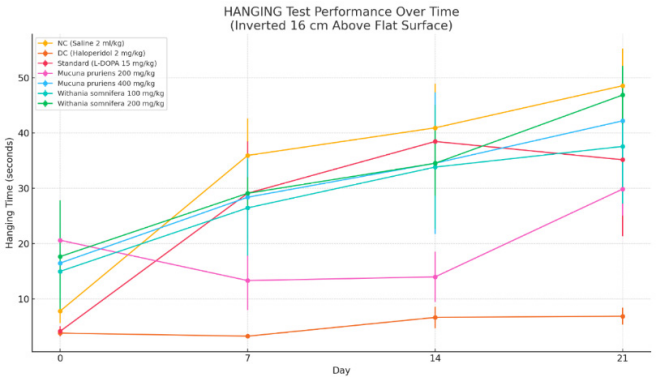


Fig. 3: Hanging Test Performance over 21 days for all treatment groups. Values are expresses as a Mean ± S.E.M/ n = 6

Neuroinflammation and Antioxidant Analysis

Figs 5–9 show the effects of treatments on oxidative stress and neuroinflammation in brain homogenates. Haloperidol significantly increased lipid peroxidation (LPO) and reduced glutathione (GSH), indicating oxidative stress, while L-DOPA, *M. pruriens*, and *W. somnifera* counteracted these effects in a dose-dependent manner, with *W. somnifera* (200 mg/kg) showing the

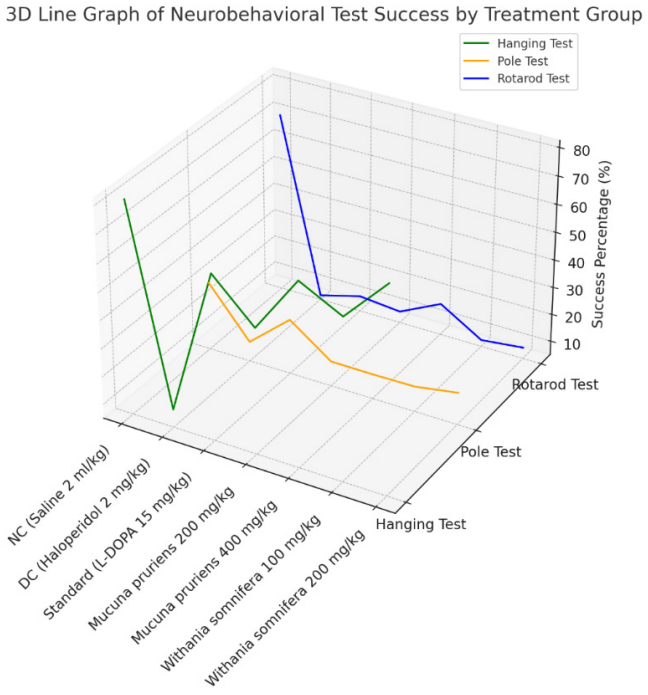


Fig. 4: Percentage of Success in Neurobehavioral Tests (Hanging, Pole, and Rotarod) for each treatment group

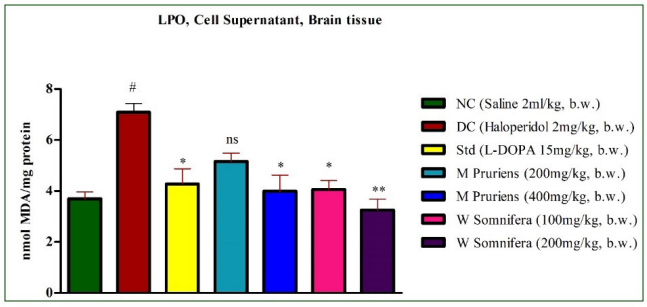


Fig. 5: LPO, Values are expressed as Mean ± S.E.M. (n = 6). # $p < 0.05$ compared to normal control; * $p < 0.05$, ** $p < 0.01$, and ns $p > 0.05$ compared to disease control (Haloperidol 2 mg/kg)

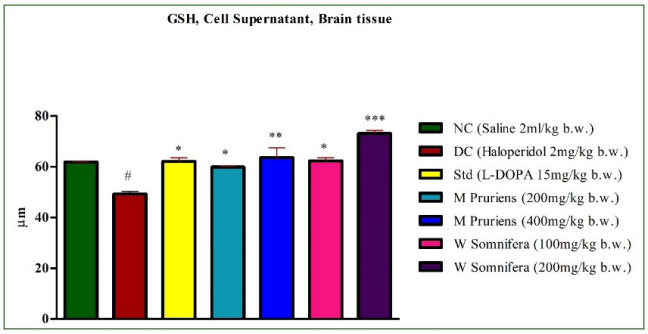


Fig. 6: GSH Values are expressed as Mean ± S.E.M. (n = 6). # $p < 0.05$ compared to normal control; * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$ compared to disease control (Haloperidol 2 mg/kg)

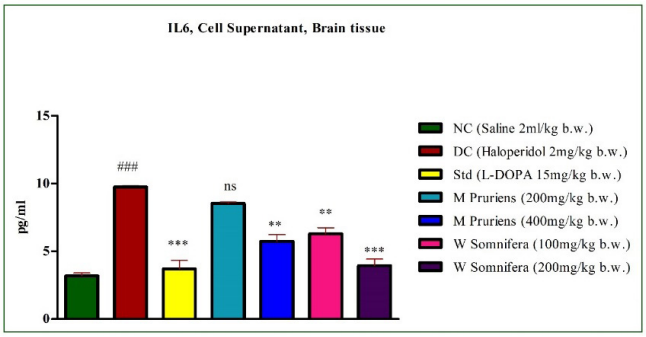


Fig. 7: Pro-inflammatory cytokines (IL6), Values are expressed as Mean ± S.E.M. (n = 6). ### $p > 0.001$ compared to normal control; *** $p > 0.001$, ** $p < 0.01$, and ns $p > 0.05$ compared to disease control (Haloperidol 2 mg/kg)

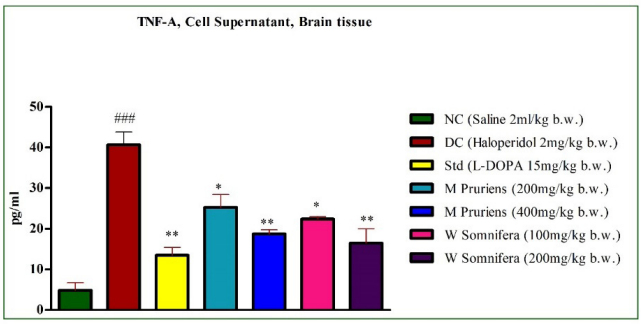


Fig. 8: Pro-inflammatory cytokines (TNF-alpha), Values are expressed as Mean ± S.E.M. (n = 6). ### $p > 0.001$ compared to normal control; * $p < 0.05$, ** $p < 0.01$ compared to disease control (Haloperidol 2 mg/kg)

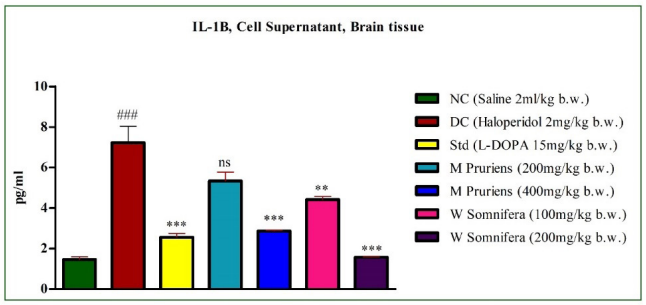


Fig. 9: Pro-inflammatory cytokines (IL-1beta), Values are expressed as Mean ± S.E.M. (n = 6). ### $p > 0.001$ compared to normal control; *** $p > 0.001$, ** $p < 0.01$, and ns $p > 0.05$ compared to disease control (Haloperidol 2 mg/kg).

strongest antioxidant activity. Haloperidol also elevated pro-inflammatory cytokines (IL-6, TNF-α, and IL-1β), reflecting neuroinflammation. Treatment with L-DOPA, *M. pruriens*, and *W. somnifera* reduced these cytokine levels dose-dependently, with *W. somnifera* (200 mg/kg) demonstrating the most pronounced anti-inflammatory and neuroprotective effects. These results highlight the potential of *W. somnifera* as a potent neuroprotective and anti-inflammatory agent.

Histopathology – Brain Tissue

Figs. 10 (A–E) illustrates the histological architecture of the hippocampus and frontal cerebellar cortex across different experimental groups. Hippocampus: The Cornu Ammonis (CA1, CA2, CA3) and dentate gyrus (DG) were clearly observed, with three layers in CA regions: pyramidal, polymorphic, and molecular. In normal/control groups, the pyramidal layer contained intact small pyramidal cells with large vesicular nuclei and basophilic cytoplasm. The molecular layer showed glial cells, neurons, and blood vessels, while the DG contained numerous ganglion cells in a single layer. Degeneration induced by haloperidol was evident as vacuolated cytoplasm, pyknotic nuclei, and thinning of the pyramidal or polymorphic layers. Treatment with *M. pruriens, W. somnifera*, and L-DOPA mitigated these changes, with the highest doses showing near-normal architecture. Cerebellar cortex: Normal groups exhibited cortical neurons with basophilic nuclei at the periphery, abundant granular cells with prominent nucleoli, and scattered oligodendrocytes. Haloperidol caused mild neuronal degeneration, vacuolations, and congested blood vessels. *M. pruriens, W. somnifera*, and standard treatments preserved cortical and granular layer architecture, reducing neuronal degeneration. No interstitial bleeding or inflammatory cell infiltration was observed in any group. H&E staining; scale bar = 200 µm.

MTT-based Cell Viability Assay

The results of the MTT-based cell viability assay indicate that various treatments cause a different cytotoxic impact on the Neuro-2a cells after 24-hour exposure (Figs 11 and 12). The untreated comparative group also exhibited very high viability of the cells levels in all the concentrations, showing good health (varying between 98.0 and 89.0%). As a positive control, haloperidol exhibited a high dose-intensity cytotoxic effect, as the cell viability decreased considerably between 78.4% at 1 µg/mL and 38.9% at 10 µg/mL. This affirms its reported neurotoxicity at improved doses and certifies the sensitivity of the test. Conversely, L-DOPA used as a negative control was lowly cytotoxic. The viability of the cells was found to be the most at 2.5 µg/mL (96.7%) and a progressive decrease until dropping back up to 82.3 at 10 µg/mL. The trend implies that L-DOPA will usually neither be harmful nor intolerable in the neuronal cells. On the same note, the *M. pruriens* had an excellent viability profile nearly identical to that of L-DOPA, with maximum viability at 97.1% (2.5 µg/mL) and over 83% at the highest dose. That implies that it is very non-toxic and may have neuroprotective characteristics. *W. somnifera* showed the same tendency, sustaining moderate, or high levels of cell viability (86.2% to 80.6%), which also proved it as a non-toxic agent and potential neuroprotectant. A toxic control DMSO 10% cell death percentage was nearly 100 (viability 0.6–0.6%), indicating the accuracy of the cell death assay and the cytopathic effect on high concentration DMSO. Together, these data emphasize that haloperidol strikingly reduces cell viability, whereas *M. pruriens and W. somnifera* possess a high cell viability feature, being good candidates as safe and neuroprotective beyond neuronal neurons.

Discussion

The present study demonstrated that both *M. pruriens* (MP) and *W. somnifera* (WS) exerted significant neuroprotective effects in a haloperidol-induced Parkinsonian model, as evidenced by improvement in behavioral motor tests, enhanced mitochondrial viability, attenuation of oxidative stress, and immunomodulation. Among them, MP at 400 mg/kg showed superior motor recovery, while WS at 200 mg/kg exhibited stronger antioxidant and anti-inflammatory responses. The findings on MP are in agreement with earlier reports highlighting its L-DOPA content as the primary contributor to dopaminergic activity.[27,28] Moreover, studies by Cili *et al*. (2017) [29] further substantiated MP as a natural source of L-DOPA, though its long-term use may lead to complications such as dyskinesia due to variability in phytoconstituents. The improvement observed in rotarod and hanging test performance in our study is consistent with these dopaminergic restorative actions. Similarly, the protective effect of WS is supported by its phytoconstituents, such as withanolides and withanosides, which are known to regulate oxidative stress and neuroinflammation.[30,31] The current findings that WS restored glutathione (GSH) and reduced lipid peroxidation are consistent with prior evidence demonstrating its role in re-establishing antioxidant enzyme systems.[32,33] Additionally, the observed suppression of IL-1β and TNF-α is in line with reports of WS exerting immunomodulatory effects in neuroinflammatory models. Histological analysis in this study further confirmed the neurorestorative potential of both extracts, showing partial neuronal recovery at higher doses. These results support the hypothesis that the combined antioxidant, dopaminergic, and anti-inflammatory activities of phytochemicals such as L-DOPA (MP) and withanolides (WS) contribute to neuronal survival. The consistency of biochemical and behavioral findings with previous literature validates the reliability of the present observations. Unlike previous works that examined these herbs individually, the present study integrates multiple experimental endpoints, including behavioral motor tests, mitochondrial viability (MTT), biochemical oxidative markers, inflammatory mediators, and histological analysis, thereby providing a comprehensive mechanistic understanding of their effects. Scientific validity is further strengthened by employing a well-established PD model and corroborating results across independent measures such as behavioral, biochemical, and cellular assays, ensuring consistency and reliability of the findings.

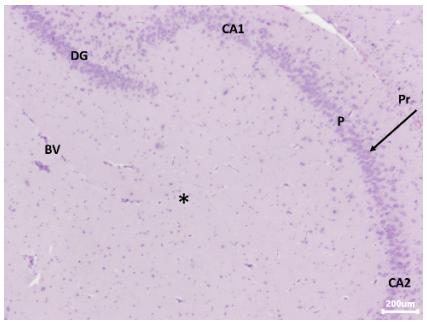
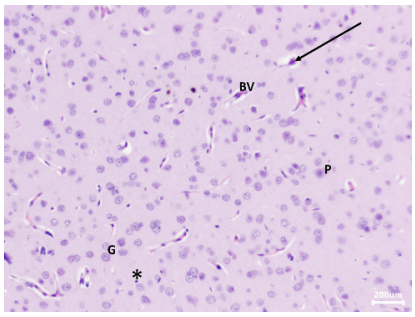
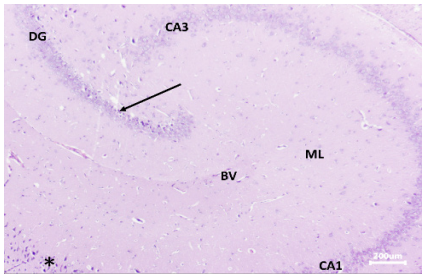
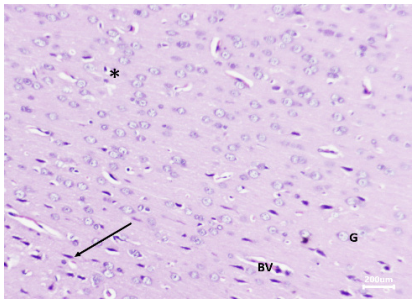
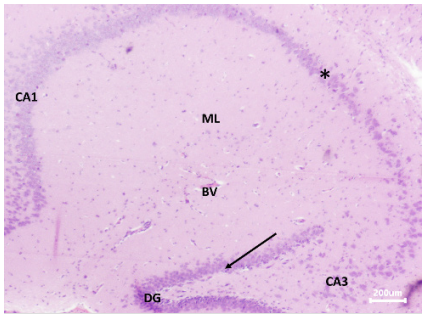
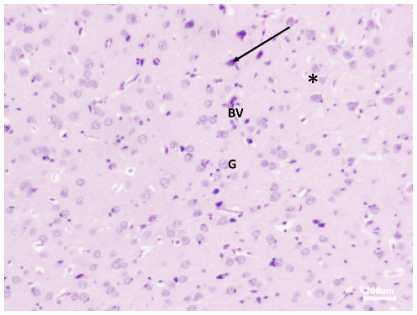
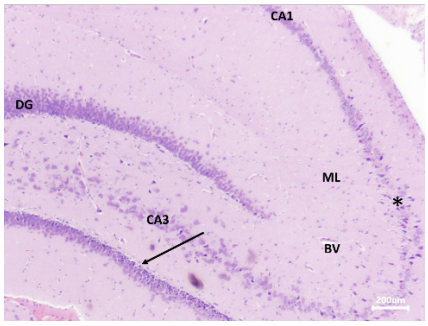
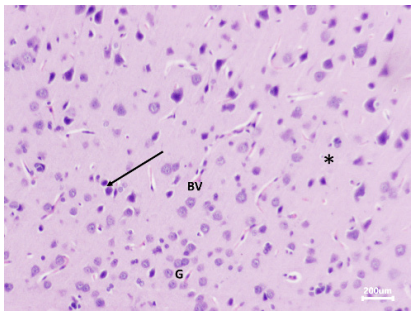
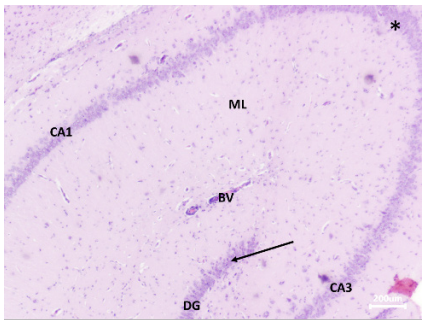
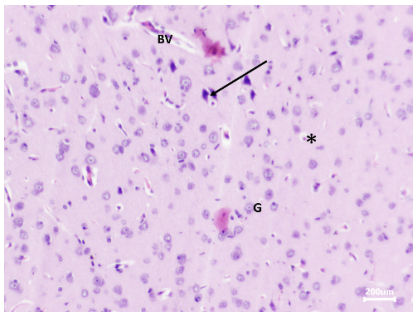
Groups	*Hippocampus tissue*	*Cerebellar cortex tissue*
Fig. 10A: Untreated Control (NC)		
Fig. 10B: Haloperidol (Positive control)		
Fig. 10C: L-DOPA (Negative control)		
Fig. 10D: *M. pruriens*		
Fig. 10E: *W. somnifera*		

Fig. 10: Brain tissue (Hippocampus and Cerebellar cortex) images of various treatments groups

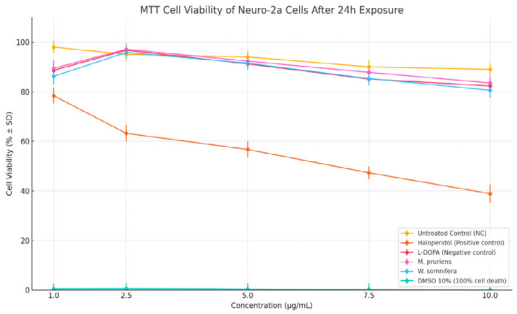


Fig. 11: MTT Cell Viability (% ± SD) in Neuro-2a Cells at Different Concentrations (24 h Exposure)

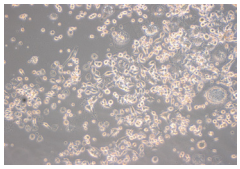
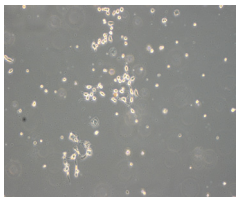
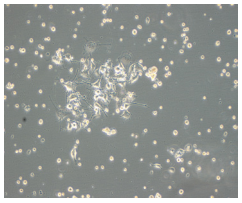
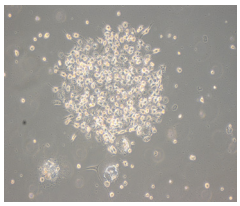
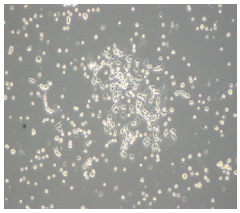
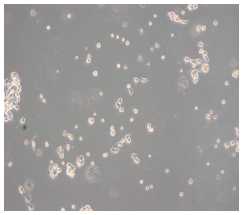
Groups	*Cell morphology*
Fig. 12A: Untreated Control (NC)	
Fig. 12B: Haloperidol (Positive control)	
Fig. 12C: L-DOPA (Negative control)	
Fig. 12D: *M. pruriens*	
Fig. 12E: *W. somnifera*	
Fig. 12F: DMSO 10 % (toxic control)	

Fig. 12: Microscopy images visually depict the effects of various treatments on Neuro-2a cell viability after 24 hours of exposure

The findings of this investigation contribute novel insights by revealing distinct but complementary neuroprotective profiles of the two herbs. MP demonstrated superior efficacy in motor recovery, primarily attributed to its natural L-DOPA content and dopaminergic restoration, whereas WS was more effective in attenuating oxidative stress and suppressing inflammatory mediators, highlighting its strong immunomodulatory role. Together, these observations suggest the possibility of complementary therapeutic application of the two plants in managing neurodegenerative disorders, particularly Parkinson's disease and drug-induced Parkinsonism. Moreover, by scientifically validating traditional ethnopharmacological claims with robust experimental evidence, this study helps bridge the gap between traditional medicine and modern pharmacology. Its comprehensive design incorporates behavioral, biochemical, cellular, and histological endpoints, and the phytopharmacological justification that aligns with previously reported mechanisms of action. The results also demonstrated dose-dependent neuroprotection, reinforcing their therapeutic potential. The findings are restricted to preclinical animal models, and translation to human conditions requires further validation. The study employed crude ethanolic extracts rather than isolating specific phytoconstituents, making mechanistic attribution less precise. In addition, the relatively short experimental duration does not allow conclusions about long-term efficacy and the absence of a direct comparison with standard PD drugs such as levodopa or selegiline limits the benchmarking of therapeutic relevance.

Conclusion

The experiment shows that *M. pruriens and W. somnifera* have neuroprotective and anti-inflammatory effects in case of haloperidol-induced neurotoxicity. Both extracts enhanced the behavioral performance, replenishment of antioxidants (GSH), lowered oxidative stress (LPO), and inhibited the production of pro-inflammatory cytokines. The most marked effects were produced with *W. somnifera* at 200 mg/kg. Their protective effect was further proved by its histopathological and *in-vitro* (MTT assay) findings. These observations suggest the possibility of these herbal extracts, especially *W. somnifera,* as a potential anti-therapeutic agent in alleviating the neurodegenerative damage and inflammation.

Acknowledgments

The authors would like to acknowledge all the scientific websites, college management, authors, and tools used to obtain and analyze the information in this study.

Conflict of Interest

None.

Funding

Self.

References

1. World Health Organization. Parkinson disease. Geneva: World Health Organization; 2023 [cited 2025 Jul 7]. Available from: https://www.who.int/news-room/fact-sheets/detail/parkinson-disease
2. World Health Organization. Parkinson disease: a public health approach. Geneva: World Health Organization; 2022 [cited 2025 Jul 7]. Available from: https://www.who.int/publications/i/item/9789240044286
3. National Institute of Neurological Disorders and Stroke. Parkinson's disease: challenges, progress, and promise [Internet]. Bethesda (MD): National Institutes of Health; 2025 Feb 25 [cited 2025 Jul 7]. Available from: https://www.ninds.nih.gov/current-research/focus-disorders/parkinsons-disease-research
4. Vaidya DB, Vaidya AB. Traditional plant-based therapies for Parkinson's disease: a review. *J Ayurveda Integr Med*. 2021;12(1):135–44. Available from: doi:10.1016/j.jaim.2019.08.005
5. Reeta KH, Mehla J, Gupta YK. Curcumin and its neuroprotective role in neurological disorders. *Neurosci Bull*. 2014;30(1):112–23. Available from doi:10.1007/s12264-013-1370-x
6. Manyam BV. Paralysis agitans and levodopa in Ayurveda: ancient Indian medical knowledge. *Mov Disord*. 1990;5(1):47–8. Available from: doi:10.1002/mds.870050111
7. Kumar H, Lim HW, More SV, *et al*. The role of free radicals in the aging brain and Parkinson's disease: convergence and parallelism. *Int J Mol Sci*. 2012;13(8):10478–504. Available from: doi:10.3390/ijms130810478
8. Kulkarni SK, Dhir A. Bacopa monniera, a nootropic drug, modulates the antioxidant and neuroprotective activity in the brain. *Phytother Res*. 2008;22(6):749–54. Available from: doi:10.1002/ptr.2364
9. Singh S, Dubey R, Sharma B. Role of natural antioxidants in the management of neurodegenerative disorders: a review. *Eur J Med Chem*. 2020;176:116113. Available from: doi:10.1016/j.ejmech.2019.116113
10. Misra A, Ghosh S, Dey CS. *Mucuna pruriens* improves Parkinsonian symptoms with better tolerability than L-DOPA in MPTP mouse model. *Phytother Res*. 2013;27(11):1692–8. Available from: doi:10.1002/ptr.4892
11. Manyam BV, Dhanasekaran M, Hare TA. Neuroprotective effects of the antiparkinson drug *Mucuna pruriens*. *Phytother Res. 2004*;18(9):706–12. Available from: doi:10.1002/ptr.1521
12. Tanner CM. Epidemiology of Parkinson's Disease. *Neurol Clin*. 1992;10(2):317–29. Available from: doi: 10.1016/S0733-8619(18)30212-3
13. Evans DA, Funkenstein MS, Albert MS, Scherr PA, Cook NR, Chown MJ, *et al*. Prevalence of Alzheimer's disease in a community population of older persons. *JAMA*. 1989;262(18):2251–6. Available from: https://pubmed.ncbi.nlm.nih.gov/2810583/
14. Kurtzke JF. Epidemiology of Amyotrophic Lateral Sclerosis. In: Rowland LP, editor. Human Motor Neuron Diseases. *Adv Neurol*. 1982;36:281–302. Available from: https://pubmed.ncbi.nlm.nih.gov/6983824/
15. Jha DK, Koneri R, Samaddar S. Antidiabetic activity of phytosaponin in STZ-induced type I diabetes in rats. *Res J Pharm Technol*. 2019;12(8):3919–26. Available from: doi:10.5958/0974-360X.2019.00675.9. Available from: https://rjptonline.org/AbstractView.aspx?PID=2019-12-8-65
16. Terry AV Jr, Gearhart DA, Warner SE, Zhang G, Bartlett MG, Middlemore ML, *et al*. Oral haloperidol or risperidone treatment in rats: temporal effects on nerve growth factor receptors, cholinergic neurons, and memory performance. *Neuroscience*. 2007;146(4):1316–32. Available from: doi:10.1016/j.neuroscience.2007.02.040
17. Bredberg E, Lennernäs H, Paalzow L. Pharmacokinetics of levodopa and carbidopa in rats following different routes of administration. *Pharm Res*. 1994;11(4):549–55. Available from: doi: 10.1023/a:1018970617104
18. Manna S, Bhattacharyya D, Mandal TK, Dey S. Neuropharmacological effects of deltamethrin in rats. *J Vet Sci*. 2006;7(2):133–6. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC3242104/
19. Hu M, Li F, Wang W. Vitexin protects dopaminergic neurons in MPTP-induced Parkinson's disease through PI3K/Akt signaling pathway. *Drug Des Devel Ther*. 2018;12:565–73. Available from: doi: 10.2147/DDDT.S156920
20. Rai SN, Yadav SK, Singh D, Singh SP. Ursolic acid attenuates oxidative stress in nigrostriatal tissue and improves neurobehavioral activity in MPTP-induced Parkinsonian mouse model. *J Chem Neuroanat*. 2016;71:41–9. Available from: doi: 10.1016/j.jchemneu.2015.12.002
21. Jollow DJ, Mitchell JR, Zampaglione NA, Gillette JR. Bromobenzene-induced liver necrosis. Protective role of glutathione and evidence for 3,4-bromobenzene oxide as the hepatotoxic metabolite. *Pharmacology*. 1974;11(3):151–69. Available from: doi: 10.1159/000136485
22. Ohkawa H, Ohishi N, Yagi K. Reaction of linoleic acid hydroperoxide with thiobarbituric acid. *J Lipid Res*. 1978;19(8):1053–7. Available from: https://pubmed.ncbi.nlm.nih.gov/103988/
23. Farombi EO, Awogbindin IO, Farombi TH, Oladele JO, Izomoh ER, Aladelokun OB, *et al*. Neuroprotective role of kolaviron in striatal redo-inflammation associated with rotenone model of Parkinson's disease. *Neurotoxicology*. 2019;73:132–41.Available from: doi: 10.1016/j.neuro.2019.03.005
24. Mir UF, Jha DK, Chatterjee S. Pro inflammatory cytokines predict the response of cancer treatment. *Indian J Pharm Educ Res*. 2025;59(2):441–52. Available from: doi:10.5530/ijper.20254089
25. Dulal TR, Jha DK, Sah SK, Fathima M. Evaluation of the antipsychotic and neuroprotective properties of *Bergenia ciliata* root extract in rat models. *Int J Pharm Sci Drug Res*. 2024;16(6):1023–31. Available from: doi:10.25004/IJPSDR.2024.160613
26. Ashhar MU, Kumar S, Ali J, Baboota S. CCRD based development of bromocriptine and glutathione nanoemulsion tailored ultrasonically for the combined anti-parkinson effect. *Chem Phys Lipids*. 2021;235:105035. Available from: doi:10.1016/j.chemphyslip.2020.105035
27. Manyam BV. An alternative medicine treatment for Parkinson's disease: *Mucuna pruriens*. *J Neurol Neurosurg Psychiatry*. 1995;60(3):307–8.
28. Pinna A, Morelli M, Carta AR, *et al*. Antiparkinsonian properties of *Mucuna pruriens* in rodent models. *Pharmacol Biochem Behav*. 1989;33(2):497–501.
29. Cilia R, Laguna J, Cassani E, *et al*. *Mucuna pruriens* in Parkinson's disease: A double-blind clinical and pharmacological study. *J Neurol Neurosurg Psychiatry*. 2017;88(12):1173–9.
30. Kulkarni SK, Dhir A. *Withania somnifera*: an Indian ginseng. *Prog Neuropsychopharmacol Biol Psychiatry*. 2008;32(5):1093–105. Available from: doi: 10.1016/j.pnpbp.2007.09.011
31. Wadhwa R, Konar A, Kaul SC. Ashwagandha as a neuroprotective agent: molecular aspects. *Indian J Med Res*. 2013;137(2):174–93.
32. Ahmad M, Saleem S, Ahmad AS, *et al*. Neuroprotective effects of *Withania somnifera* on oxidative stress–induced neuronal damage in rats. *Phytother Res*. 2005;19(12):1076–82.
33. Majolo F, Colpo AC, da Silva AM, *et al*. Neuroprotective mechanisms of *Withania somnifera*: A systematic review. *Phytomedicine*. 2021;91:153694.

HOW TO CITE THIS ARTICLE: Ogwang MB, Jha DK, Chatterjee S. Comparative Study of Neuroprotective Activity of *Mucuna pruriens* and *Withania somnifera* Against Haloperidol-Induced Parkinson's Disease in an Animal Model. Int. J. Pharm. Sci. Drug Res. 2025;17(5):452-460. **DOI:** 10.25004/IJPSDR.2025.170505