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Neuroprotective Assessment of Putranjiva Roxburghii Leaves Extract in Scopolamine-Induced Model in Rats

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ABSTRACT: The study aimed to evaluate the antioxidant, neuroprotective, and memory-enhancing potential of the methanolic extract of Putranjiva roxburghii leaves against scopolamine
induced cognitive impairment in Wistar rats.
Method: The leaves were extracted using methanol and evaluated for phytochemical constituents, total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity by the DPPH assay. Acute oral toxicity was assessed according to OECD Guideline 423. Neuroprotective activity was evaluated at doses of 250 and 500 mg/kg using the Elevated Plus Maze (EPM) and Morris Water Maze (MWM), with donepezil as the standard drug.
Result: The extract yield was 7.942%, with TPC and TFC values of 29.65 mg GAE/g and 22.69 mg RE/g, respectively. The extract exhibited concentration dependent antioxidant activity and no acute toxicity up to 2000 mg/kg. Both doses improved learning and memory reduced lipid peroxidation, enhanced antioxidant defenses, and attenuated neuronal degeneration. The 500 mg/kg dose showed greater protective effects than the 250 mg/kg dose.
Conclusion: The methanolic extract of Putranjiva roxburghii leaves demonstrated promising antioxidant and neuroprotective effects against scopolamine-induced cognitive impairment. Further studies are required to establish its active constituents, mechanisms of action, and therapeutic potential.

1. INTRODUCTION

Neurodegenerative disorders are mainly defined by the gradual loss of neurons, resulting in the slow decline of cognitive and motor abilities. Among these disorders, Alzheimer's disease (AD) and Parkinson's disease (PD) are the most common, having a significant effect on aging populations globally. Current pharmacological interventions primarily aim to alleviate symptoms rather than prevent disease progression. This limited effectiveness is largely due to the blood-brain barrier (BBB), which hinders the passage of therapeutic agents into the central nervous system. Additionally, the pathophysiological processes associated with many neurodegenerative diseases are still not well understood. As a result, researchers have begun to investigate environmental factors such as exposure to heavy metals, pesticides, and air pollutants as possible contributors to neurodegeneration.

The implementation of neural-regeneration strategies aiming at reconstructing neuronal and synaptic networks holds potential as a therapeutic approach for AD. Neurogenesis, characterized by neurite outgrowth, is one of the neural-regeneration processes crucial for this purpose. It involves the branching of neurites, subsequent axonal, and dendritic elongation in maturing neurons. This fundamental process plays a vital role in constructing functional neuronal networks and is regarded as a hallmark of neuronal differentiation. Neurite outgrowth serves as a crucial initial step in

the formation of the neuronal network. Therefore, drug discovery and development efforts targeted at promoting neurite outgrowth are an essential for understanding molecular mechanisms and developing effective treatments for axonal and synaptic damages

Putranjiva roxburghii Wall. (Family Putranjivaceae) is an evergreen tree found growing in wild as well as cultivated in the Indian subcontinent. It is commonly called Putranjiva, is a deciduous, evergreen tree of about 18m tall having grey bark. Leaves elliptic-oblong to ovate-lanceolate, unequal sided at the base, dark green and shining inappearance. Flowers small male in dense,rounded clusters, yellowish in colour,female flowers solitary or 2-3 together, green. Drupes ellipsoid or globose and white-tomentose, stone pointed, rugose, very hard and normally having a single seed. It is regarded as one of the best herbs for rejuvenation and restoration of the female reproductive system in the Indian traditional system of medicine. Apart from having analgesic, antipyretic, and anti-inflammatory activities. It is also used for treating azoospermia, elephantiasis, eye infection, habitual abortion, sterility, constipation, cough, cold and fever.

The study aimed to investigate the identification of Putranjiva roxburghii Leaves extracts, elucidating their phytochemical profiles. Moreover, the present study also evaluated the neuroprotective and anti-Alzheimer's potential of Putranjiva roxburghii Leaves extract using a scopolamine-induced amnesia model in rats, and to scientifically validate its traditional medicinal claims for the management of cognitive dysfunction and Alzheimer's disease.

MATERIAL AND METHODS

Collection and Identification of Plant Material

For the present study, fresh leaves of Putranjiva roxburghii were collected from a local region of Bhopal, Madhya Pradesh, India. The plant material was authenticated by a qualified botanist for correct taxonomic identification. After collection, the leaves were thoroughly washed with running water to remove dust, soil particles, and other extraneous impurities.

The cleaned plant material was then shade-dried at room temperature for 5–7 days, followed by controlled oven drying at 45 °C until a constant weight was obtained to ensure complete removal of moisture. The fully dried leaves were subsequently coarsely powdered using a mechanical grinder under hygienic conditions. The powdered material was passed through a suitable sieve to ensure uniform particle size and stored in airtight containers to protect it from moisture and contamination. Approximately 350 g of the powdered leaves material was obtained and used for extraction and further pharmacological evaluation, including neuroprotective studies.

Preparation of Extract: Drying, Powdering, and Extraction of Plant Material

Fresh leaves of Putranjiva roxburghii were collected, shade-dried, and coarsely powdered using a mechanical grinder. The powdered material was accurately weighed and transferred into a Whatman filter paper thimble, which was placed in the extraction chamber of a Soxhlet apparatus. Approximately 500 mL of methanol was used as the extraction solvent and poured into a clean round-bottom flask connected to the Soxhlet apparatus. The extraction was carried out by heating the solvent on a heating mantle, allowing methanol to boil, evaporate, condense through the condenser, and continuously percolate through the powdered plant material. The solvent siphoned back into the boiling flask after each extraction cycle, carrying the dissolved phytoconstituents. This process was continued for 6–8 hours until the siphoning solvent became nearly colorless, indicating complete extraction of the bioactive constituents. After completion of the extraction, the methanolic extract was filtered to remove any insoluble plant debris. The filtrate was concentrated under reduced pressure using a rotary vacuum evaporator at a controlled temperature to remove the solvent without affecting the thermolabile constituents. The concentrated extract was further dried to obtain a semi-solid crude methanolic extract. The percentage yield of the extract was calculated based on the initial weight of the dried plant material. The dried methanolic extract was transferred to airtight amber-colored glass containers and stored at 4–8°C until further use for phytochemical analysis and pharmacological studies.

Preliminary Phytochemical Screening of Putranjiva roxburghii LeavesExtract

The crude extract of Putranjiva roxburghii leaves was subjected to systematic qualitative phytochemical screening to detect the presence of major secondary metabolites such as alkaloids, flavonoids, tannins, saponins, glycosides, phenols, steroids, terpenoids, proteins, amino acids, and carbohydrates. Standard procedures were followed as per established phytochemical protocols.

Quantitative Phytochemical Screening

Total Phenolic Content (TPC)

A known volume of Putranjiva roxburghii leaves extract was taken in a test tube and mixed with Folin–Ciocalteu reagent. After a short interval, sodium carbonate solution (7.5%) was added to neutralize the reaction mixture. The solution was then incubated at room temperature for 30–60 minutes in dark conditions to ensure complete color development. The absorbance of the resulting blue-colored solution was recorded at 760 nm using a UV–Visible spectrophotometer against a blank. Gallic acid was used to prepare the calibration curve. Total phenolic content was expressed as mg of gallic acid equivalents per gram of extract (mg GAE/g)

Total Flavonoid Content (TFC)

A measured amount of plant extract was mixed with 10% aluminium chloride solution, potassium acetate, and distilled water in a test tube. The mixture was shaken well and incubated at room temperature for 30 minutes to allow complete complex formation. The absorbance of the developed yellow color was measured at 510 nm using a UV–Visible spectrophotometer against a blank. Rutin was used for calibration curve preparation. Total flavonoid content was expressed as mg of Rutin equivalents per gram of extract (mg RE/g).

Assessment of In-vitro Antioxidant Activity of Putranjiva roxburghii leaves extract

DPPH Free Radical Scavenging Assay

A 0.1 mM DPPH solution was freshly prepared in methanol and protected from light. The Putranjiva roxburghii leaves extract was dissolved in methanol to prepare a stock solution, from which different concentrations (25, 50, 100, 200, and 400 µg/mL) were prepared.One milliliter of each concentration of the extract was mixed with 1 mL of the DPPH solution and vortexed thoroughly. The reaction mixtures were incubated at room temperature in the dark for 30 minutes to ensure complete reaction between the antioxidants and DPPH radicals. The absorbance of each reaction mixture was measured at 517 nm using a UV–Visible spectrophotometer. The antioxidant activity of the extract was determined by comparing the absorbance of the sample with that of the control.

Acute Oral Toxicity Study of Putranjiva roxburghii leaves extract

An acute oral toxicity study of Putranjiva roxburghii leaves extract was conducted in accordance with the OECD Guideline 423 (Acute Toxic Class Method). Healthy adult female Wistar rats were used for the study after overnight fasting, with free access to water. The extract was administered orally as a single dose at 5, 50, 300, and 2000 mg/kg body weight. Following administration, the animals were observed individually during the first 30 minutes, periodically during the first 24 hours (with special attention during the first 4 hours), and thereafter daily for 14 days. The animals were monitored for signs of toxicity, including:

Changes in skin, fur, eyes, and mucous membranes, Alterations in behavior and locomotor activity, Tremors, convulsions, salivation, diarrhea, lethargy, sleep, and coma

In-vivo Assessment of Anti-Amnesia Activity (Scopolamine-Induced Model)

Scopolamine induced amnesia model

Healthy adult Wistar rats were used in the study and maintained under standard laboratory conditions with free access to food and water, following IAEC and CPCSEA guidelines. Scopolamine hydrobromide was used to induce amnesia and was freshly prepared in normal saline. Animals in the inducer group received scopolamine at a dose of 1 mg/kg body weight, orally (p.o.), administered 30–60 minutes before behavioral testing to induce cognitive impairment by disrupting cholinergic neurotransmission. Memory and learning were evaluated using standard behavioral models such as the Elevated plus Maze and Morris Water Maze test. The test drug was given orally to the treatment group, beginning on the 6th day after scopolamine administration, and continued for 14 days.

Animals were divided into 5 groups (n=6 per group)

Group	Description	Treatment
Group I	Normal Control	Normal saline treated group (received normal saline 5 mL/kg body weight orally)
Group II	Negative Control	Scopolamine-treated group (received scopolamine 1 mg/kg, p.o.) to induce amnesia
Group III	Standard Drug Group	Standard treatment group (received Donepezil 1.5 mg/kg, p.o. + Scopolamine 1 mg/kg, p.o.)
Group IV	Putranjiva roxburghii Leaves ExtractLow Dose	Low-dose treatment group (received methanolic Leavesextract 250 mg/kg body weight, p.o. + Scopolamine 1 mg/kg, p.o.)
Group V	Putranjiva roxburghii Leaves ExtractHigh Dose	High-dose treatment group (received methanolic Leaves extract 500 mg/kg body weight, p.o. + Scopolamine 1 mg/kg, p.o.)

Evaluation of Neuroprotective Activity by behavioural Assessment

Rats are divided into different groups and pretreated with the extract, followed by the induction of amnesia using scopolamine. The anti-amnesic effect of the extract is evaluated using appropriate behavioural models.

Behavioural Studies

Behavioural testing was performed during the light phase and at consistent times. Animals were habituated to the testing room 30 min before testing. The investigator conducting behavioural tests was blinded to group allocation.

A. Elevated plus maze (EPM)

The Elevated plus Maze (EPM) was used to assess anxiety, exploration, and learning and memory in experimental animals. The apparatus consisted of two open arms and two closed arms (49 × 10 cm), with closed arms having 40 cm high walls, elevated 50 cm above the ground. On the 10th day, 60 minutes after drug administration, each mouse was placed at the end of an open arm facing away from the central platform, and the time taken to enter any closed arm was recorded as transfer latency (TL), representing learning acquisition. If the mouse failed to enter a closed arm within 180 seconds, it was gently guided into one, and TL was recorded as 180 seconds. Animals were allowed to explore the maze for 15 seconds before returning to their home cages. On the 14th day, TL was again recorded to assess memory retention, and the maze was cleaned with 30% ethanol between sessions to eliminate olfactory cues**.**

B. Morris water maze

The Morris Water Maze test was used to assess learning and spatial memory in experimental animals. The apparatus consisted of a circular pool filled with opaque water maintained at 22 ± 2°C and divided into four quadrants using visual cues, with a submerged platform fixed 1 cm below the water surface in the target quadrant. On day 10 of drug administration, animals were acclimatized by swimming for 120 seconds without the platform, followed by four consecutive days of training with four trials per day. Escape latency time (ELT) to locate the platform was recorded for each trial, with a maximum cut-off time of 120 seconds. On day 14, the platform was removed for the probe trial, and animals were allowed to swim for 60 seconds; the time taken to reach the target quadrant was recorded to evaluate memory retention.

RESULTS AND DISCUSSION

Percentage Yield

The methanolic extract of Putranjiva roxburghii leaves exhibited a percentage yield of 7.942%, with 27.8 g of crude extract. The observed yield indicates that methanol is an efficient solvent for extracting polar phytoconstituents from the plant material. The

extraction yield may be attributed to the presence of bioactive compounds such as phenolics, flavonoids, tannins, glycosides, and alkaloids.

Table 1: Percentage Yield of plant material

Plant name	Solvent	Theoretical weight	Yield(gm)	% yield
Putranjiva roxburghii leaves	Methanol	350	27.8	7.942%

Qualitative Phytochemical estimation

Table 2: Phytochemical test of Putranjiva roxburghii leaves extract of Methanolic extract

S.No.	Phytoconstituents	Test Performed	Observation	Result
1	Alkaloids	Dragendorff's Test	Orange-red precipitate formed	+ Present
2	Alkaloids	Mayer's Test	Cream-colored precipitate formed	+ Present
3	Flavonoids	Shinoda Test	Pink to crimson-red coloration	+ Present
4	Flavonoids	Alkaline Reagent Test	Intense yellow color disappeared on addition of dilute acid	+ Present
5	Phenolic Compounds	Ferric Chloride Test	Bluish-green coloration observed	+ Present
6	Tannins	Ferric Chloride Test	Blue-black coloration developed	+ Present
7	Saponins	Foam Test	Persistent froth observed for 15 min	+ Present
8	Glycosides	Keller–Killiani Test	Brown ring formed at the interface	+ Present
9	Terpenoids	Salkowski Test	Reddish-brown ring at the interface	+ Present
10	Steroids	Liebermann–Burchard Test	Bluish-green coloration developed	+ Present
11	Carbohydrates	Molisch's Test	Violet ring formed at the interface	+ Present
12	Reducing Sugars	Fehling's Test	Brick-red precipitate formed	+ Present
13	Proteins	Biuret Test	No violet coloration observed	– Absent
14	Amino Acids	Ninhydrin Test	No purple coloration observed	– Absent

Quantitative Analysis

Total Phenolic content (TPC) estimation

Total Phenolic Content in Putranjiva roxburghii leaves extract

Table 3: Total Phenolic Content

S. No.	Absorbance	Mean±SD	TPC in mg/gm equivalent of Gallic acid
1.	0.186	0.197±0.011015	29.65 mg/gm
2.	0.198		
3.	0.208		

The total phenolic content (TPC) of the methanolic extract of Putranjiva roxburghii leaves was determined using the Folin–Ciocalteu colorimetric method, with gallic acid as the reference standard. A standard calibration curve was prepared using gallic acid at concentrations of 10–90 µg/mL, the findings demonstrate that the methanolic extract of Putranjiva roxburghii leaves is a rich source of phenolic compounds. The presence of 29.65 mg GAE/g of total phenolics provides scientific evidence supporting the antioxidant potential and therapeutic value of the plant. These phenolic constituents are likely to contribute significantly to the biological activities of the extract and justify its further evaluation for antioxidant and other pharmacological applications.

Total Flavonoids content (TFC) estimation

Total Flavonoid Content in Putranjiva roxburghii leaves extract

Table 4: Total Flavonoid Content

S. No.	Absorbance	Mean±SD	TFC in mg/gm equivalent of Rutin
1.	0.158	0.163±0.007371	**22.69mg/gm**
2.	0.161		
3.	0.172		

The total flavonoid content (TFC) of the methanolic extract of Putranjiva roxburghii leaves was determined using the aluminium chloride colorimetric method, with rutin as the reference standard. A calibration curve was prepared using rutin at concentrations ranging from 10–90 µg/mL, which showed a gradual increase in absorbance with increasing concentration, confirming the linearity and suitability of the method for flavonoid quantification. Total flavonoid content of 22.69 mg RE/g confirms that the methanolic extract of Putranjiva roxburghii leaves is a valuable source of flavonoids with significant therapeutic potential. The combined presence of appreciable levels of flavonoids and phenolic compounds provides strong scientific evidence for the antioxidant capacity of the extract and supports its further investigation for various pharmacological and medicinal applications.

Anti-Oxidant Activity

DPPH 2, 2- diphenyl-1-picryl hydrazyl Assay

Table 5: DPPH radical scavenging activity of Methanol extract of Putranjiva roxburghii leaves

Concentration (µg/mL)	Absorbance	% Inhibition
20	0.536	45.967
40	0.471	52.520
60	0.389	60.786
80	0.301	69.657
100	0.224	77.419
Control		**0.992**
IC50		**31.83**

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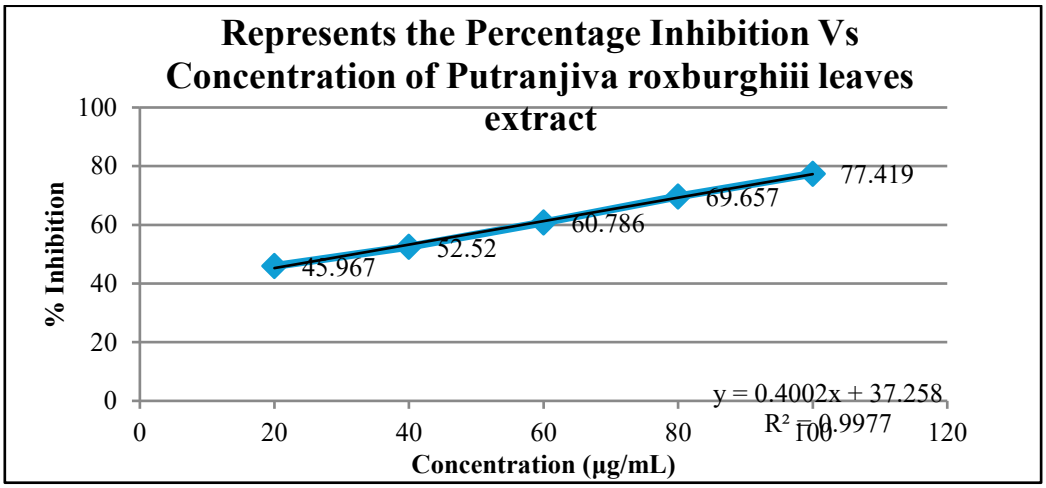


Figure 1: Represents the Percentage Inhibition Vs Concentration of Putranjiva roxburghiileaves extract

The standard ascorbic acid exhibited a significant concentration-dependent increase in DPPH radical scavenging activity. At a concentration of 20 µg/mL, ascorbic acid showed 47.13% inhibition, which increased gradually with increasing concentration and reached 55.47%, 64.32%, 72.66%, and 81.20% inhibition at concentrations of 40, 60, 80, and 100 µg/mL, respectively. The calculated IC_{50} value of ascorbic acid was 26.81 µg/mL, indicating its strong antioxidant capacity. The lower IC_{50} value reflects higher scavenging efficiency, as a smaller amount of standard compound is required to reduce 50% of DPPH radicals. The progressive increase in inhibition with increasing concentration confirms the dose-dependent antioxidant behavior of ascorbic acid.

The methanolic extract of Putranjiva roxburghii leaves also demonstrated notable DPPH radical scavenging activity. The extract showed 45.96% inhibition at 20 µg/mL, which increased progressively to 52.52%, 60.78%, 69.65%, and 77.41% at concentrations of 40, 60, 80, and 100 µg/mL, respectively.

Acute Oral Toxicity Study of Putranjiva roxburghii leaves extract

Table 6: Observations of Acute Oral Toxicity Study

Parameter Observed	5 mg/kg	50 mg/kg	300 mg/kg	2000 mg/kg
Mortality	Absent	Absent	Absent	Absent
Changes in Skin and Fur	Normal	Normal	Normal	Normal
Eyes and Mucous Membranes	Normal	Normal	Normal	Normal
Tremors	Absent	Absent	Absent	Absent
Convulsions	Absent	Absent	Absent	Absent
Salivation	Absent	Absent	Absent	Absent
Locomotor Activity	Normal	Normal	Normal	Normal
Behavioral Changes	Absent	Absent	Absent	Absent
Lethargy	Absent	Absent	Absent	Absent
Sleep/Coma	Absent	Absent	Absent	Absent
Food Intake	Normal	Normal	Normal	Normal
Water Intake	Normal	Normal	Normal	Normal

Body Weight Changes	Normal Increase	Normal Increase	Normal Increase	Normal Increase
Gross Pathological Changes	Absent	Absent	Absent	Absent

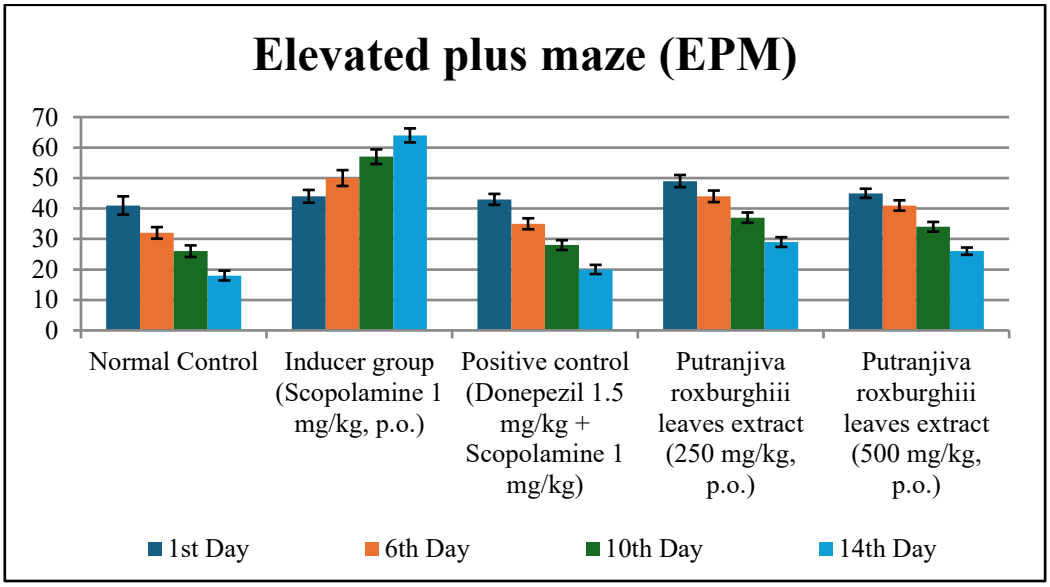
The findings of the acute oral toxicity study demonstrate that the methanolic extract of Putranjiva roxburghii leaves is non-toxic and safe following single-dose oral administration up to 2000 mg/kg body weight. The absence of mortality, clinical signs of toxicity, behavioral abnormalities, and gross pathological changes confirm the excellent safety profile of the extract. These results provide a strong basis for selecting appropriate doses for subsequent pharmacological studies and support the potential therapeutic use of Putranjiva roxburghii leaves in herbal medicine.

Analysis of general parameters

Behavioral analysis

Table 7: Elevated plus maze (EPM)

Group	1st Day	6th Day	10th Day	14th Day
Normal Control	41 ± 3.0	32 ± 1.9	26 ± 1.9	18 ± 1.6
Inducer group (Scopolamine 1 mg/kg, p.o.)	44 ± 2.1	50 ± 2.6	57 ± 2.4	64± 2.3
Positive control (Donepezil 1.5 mg/kg + Scopolamine 1 mg/kg)	43 ± 1.8	35 ± 1.8	28 ± 1.6	20 ± 1.5
Putranjiva roxburghii leaves extract (250 mg/kg, p.o.)	49± 2.0	44± 1.9	37± 1.7	29 ± 1.6
Putranjiva roxburghii leaves extract (500 mg/kg, p.o.)	45 ± 1.5	41 ± 1.7	34 ± 1.6	26 ± 1.2



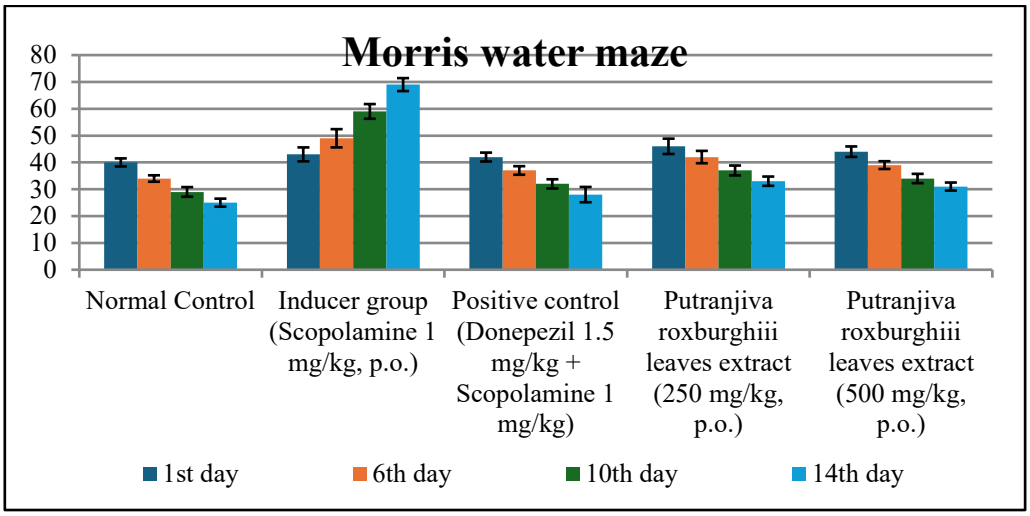
Graph 1: Behavioural Analysis of Elevated plus maze (EPM)

The behavioral performance of experimental animals was evaluated using the Elevated Plus Maze (EPM) test, which is a well-established model for assessing learning and memory The normal control group exhibited a progressive reduction in transfer latency from 41 ± 3.0 seconds on Day 1 to 18 ± 1.6 seconds on Day 14, indicating normal learning ability and memory consolidation. In contrast, the scopolamine-treated group showed a gradual increase in transfer latency from 44 ± 2.1 seconds to 64 ± 2.3 seconds, demonstrating significant impairment of learning and memory.

The positive control group treated with Donepezil (1.5 mg/kg) along with scopolamine showed a marked improvement in cognitive performance. Transfer latency progressively decreased from 43 ± 1.8 seconds on Day 1 to 20 ± 1.5 seconds on Day 14, approaching the values of the normal control group. Donepezil, an acetylcholinesterase inhibitor, enhances cholinergic neurotransmission by preventing the breakdown of acetylcholine. Although the improvement was slightly lower than that produced by Donepezil, the extract exhibited a clear dose-dependent nootropic effect, with the 500 mg/kg dose showing superior efficacy compared to 250 mg/kg.

Table 8: Morris water maze

Group	1st day	6th day	10th day	14th day
Normal Control	40± 1.5	34 ± 1.2	29 ± 1.8	25 ± 1.5
Inducer group (Scopolamine 1 mg/kg, p.o.)	43± 2.6	49± 3.4	59 ± 2.74	69 ± 2.42
Positive control (Donepezil 1.5 mg/kg + Scopolamine 1 mg/kg)	42± 1.66	37± 1.6	32 ± 1.71	28 ± 2.86
Putranjiva roxburghii leaves extract (250 mg/kg, p.o.)	46 ±2.86	42 ± 2.3	37 ± 1.84	33± 1.73
Putranjiva roxburghii leaves extract (500 mg/kg, p.o.)	44±1.95	39 ± 1.45	34 ± 1.73	31 ± 1.5



Graph 2: Behavioural Analysis of Morris water maze

The Morris Water Maze (MWM) test was employed to evaluate spatial learning and memory in experimental rats. The methanolic extract of Putranjiva roxburghii leaves was compared with the standard drug Donepezil. The normal control group demonstrated progressive learning throughout the study, with escape latency decreasing from 40 ± 1.5 seconds on Day 1 to 25 ± 1.5 seconds on Day 14, indicating normal spatial memory acquisition and retention. In contrast, the scopolamine-treated group exhibited a marked increase in escape latency from 43 ± 2.6 seconds to 69 ± 2.42 seconds, confirming severe impairment of learning and memory.The positive control group treated with Donepezil (1.5 mg/kg) in combination with scopolamine showed significant improvement in cognitive performance. Escape latency progressively declined from 42 ± 1.66 seconds on Day 1 to 28 ± 2.86 seconds on Day 14, indicating effective reversal of scopolamine-induced amnesia. Animals receiving 250 mg/kg of the extract showed a gradual reduction in escape latency from 46 ± 2.86 seconds on Day 1 to 33 ± 1.73 seconds on Day 14. Similarly, animals treated with 500 mg/kg exhibited a greater improvement, with escape latency decreasing from 44 ± 1.95 seconds to 31 ± 1.5 seconds during the study period.

CONCLUSION

The present study provides comprehensive scientific evidence supporting the neuroprotective potential of the methanolic extract of Putranjiva roxburghii leaves against scopolamine-induced memory impairment in experimental rats. The extract was

found to be rich in biologically active phytoconstituents, including phenolics, flavonoids, alkaloids, tannins, glycosides, terpenoids, and saponins, which collectively contribute to its therapeutic properties. The extract exhibited significant antioxidant activity in the DPPH assay and demonstrated an excellent safety profile. Behavioral investigations using the Elevated Plus Maze and Morris Water Maze clearly established that the methanolic extract significantly improved learning and memory in scopolamine-treated animals. Among the two doses investigated, the 500 mg/kg dose consistently produced superior pharmacological effects compared with the 250 mg/kg dose and exhibited efficacy approaching that of the standard drug donepezil.

In conclusion, the present investigation establishes that Putranjiva roxburghii leaves possess significant antioxidant, neuroprotective, and nootropic properties. The extract effectively protects neuronal tissue against scopolamine-induced oxidative damage, improves cognitive function, and preserves brain histoarchitecture. Therefore, Putranjiva roxburghii may serve as a promising natural source for the development of novel herbal therapeutics for the prevention and management of Alzheimer's disease and other neurodegenerative disorders.

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Competing interest statement

The authors declare that they have no conflicts of interest.

Author contribution

Dheeraj Kumar Gupta conceptualized and designed the study, conducted the experimental work, performed data acquisition and analysis, and drafted the manuscript. Dr. Priyanka Namdeo* provided supervision throughout the research process, contributed to the study design and interpretation of data.

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