RESEARCH PAPER

Integrated Molecular Docking and In Vitro Neuroprotective Assessment of p-Cymene in SH-SY5Y Cells

Lalit Parihar[1,*], Sanjar Alam[2], Ajay Pal Singh[1]
[1]Lingaya's Vidyapeeth (Deemed to be University), Faridabad, Haryana-121002
[2]R.V. Northland Institute, Dadri, G. B. Nagar Uttar Pradesh India
***Corresponding author: Lalit Parihar, Research Scholar, School of Pharmacy, Lingaya's Vidyapeeth (A Deemed-to-be-University), Nachauli, Jasana Road, Faridabad, Haryana, India.**
Email: lalitpparihar@gmail.com
Received: 05th July, 2026; **Revised:** 15th July, 2026; **Accepted:** 22nd July, 2026; **Available Online:** 28th July, 2026

ABSTRACT
Natural bioactive compounds continue to attract scientific attention due to their safety, accessibility, and broad therapeutic potential. Among these, monoterpenes present in essential oils have shown promising pharmacological activities, including neuroprotective effects. The present study aimed to evaluate the neuroprotective potential of p-cymene, a widely distributed monoterpene, using in silico molecular docking and in vitro neuroprotective assessment in SH-SY5Y cells. Molecular docking was performed using AutoDock against selected target proteins to understand binding affinity and interaction patterns. The docked phytochemicals exhibited binding energies ranging from −6.4 to −9.6 kcal/mol, indicating favorable interactions primarily mediated through van der Waals and alkyl forces, suggesting potential modulation of protein activity through non-covalent interactions. For experimental validation, neuroprotection was assessed using the MTT assay in SH-SY5Y cells exposed to increasing concentrations of p-cymene (1–10 mg/mL). The results demonstrated a clear dose-dependent response. Lower concentrations to medium concentrations (1 and 7 mg/mL) showed increased cell viability compared to control, indicating possible enhancement of mitochondrial activity or adaptive cellular responses. However, a slight decline in cell viability was observed at higher concentrations, with slightly lesser neuroprotective effects at 10 mg/ml. Overall, the study highlights the biphasic biological behavior of p-cymene, characterized by non-toxic or mildly stimulatory effects at low concentrations and pronounced cytotoxicity at higher doses. These findings support the potential of p-cymene as a bioactive compound with concentration-dependent therapeutic relevance, warranting further mechanistic and preclinical investigations.
Keywords: p-Cymene; Monoterpenes; Cytotoxicity; MTT assay; SH-SY5Y cells; Molecular docking; Cell viability; Natural bioactive compounds; Dose-dependent response.
How to cite this article: Parihar L, Alam S, Singh AP. Integrated Molecular Docking and In Vitro Neuroprotective Assessment of p-Cymene in SH-SY5Y Cells. Int J Drug Deliv Technol. 2026;16(63s):25-33. DOI: 10.25258/ijddt.16.63s.4
Source of support: Nil.
Conflict of interest: None

1. Introduction

Medicinal plants have long been considered a rich source of natural bioactive compounds due to their relatively cheap, easily available, and generally non-toxic nature. Because of this, they are often integrated into traditional systems of healthcare [1]. It is estimated that close to 75% of the world population depends, either directly or indirectly, on plant-based therapies for various critical disorders. A WHO study estimated that almost 21,000 plant species may have potential medicinal use. The huge diversity represented continues to drive research efforts for the discovery of useful phytochemicals from various plant sources [2].

Compared to synthetic drugs, medicines of plant origin are often considered safer, with fewer toxicities. In neuroprotective therapy, the harmful side effects of conventional synthetic drugs have motivated scientists to explore other neuroprotective agents from natural sources [3]. Various investigations have pointed out that bioactive phytochemicals can inhibit key stages in the progression of neurodegeneration. Some phytochemicals have already been approved by the FDA as active ingredients in drugs for neuronal activity, which proves their pharmacological significance. Therefore, natural compounds continue to be a fascinating prospect for the design of novel neuroprotective drugs [4,5].

Monoterpenes serve key functions in the fragrance quality of many plants and are major constituents of essential oils. Due to their impact on the VOC emission profiles, mediating interactions between

plants and insects, these chemicals also contribute to complex ecological interactions beyond their sensory function [6]. By directly and indirectly modulating levels of ROS, monoterpenes also support plants' responses to oxidative stress. By modulating photosynthetic efficiency and partitioning of metabolic resources, they also enable plants to respond to a range of abiotic stresses [7].

Although plants are the main source of monoterpenes, mammals and microbes can also produce them. Saccharomyces cerevisiae has been engineered to produce geraniol, while other engineered microbial systems such as Escherichia coli have also been developed for limonene production [8]. Some insects naturally synthesize monoterpenes; for example, honeybees secrete certain monoterpenes from their mandibular glands to facilitate in-hive signaling, while the pine bark beetles use them for pheromonal communication. Monoterpenes are the structural derivatives of geranyl diphosphate and are synthesized through methods such as isomerization and cyclization. They are isoprenoids, consisting of two joined isoprene units and 10 carbon atoms [9].

Monoterpenes are a class of compounds showing a wide range of pharmacological properties, such as analgesic, anti-inflammatory, neuroprotective and anticancer actions. Further bioactivities as antifungal, antibacterial, antioxidant, antispasmodic, and vasodilatory properties are highlighted in recent scientific studies. Moreover, experimental investigations point to potential neuroprotective properties, such as antidepressant and anxiolytic advantages, as well as memory improvements in models of dementia and Parkinson's disease [10]. Several monoterpenes, including borneol, camphor, geraniol, pinene, and thymol, have generated particular interest because of their promising therapeutic potential across a wide range of medical specialties. P-cymene is one of the most widespread monoterpenes, occurring in more than 100 plant species. It is commonly used as an active ingredient in various fungicides and insecticides and as a flavoring in the food industry. The JECFA has established a toxicological threshold of 1800 µg per person per day for p-cymene, belonging to structural class I, which describes effective metabolism, a simple chemical structure, and minimal oral toxicity. Besides that, a safety assessment performed in 2021 did not show any evidence for human genotoxicity. IFRA classes p-cymene as non-toxic, non-persistent, and non-bio accumulative [11,12].

A crucial regulatory enzyme in the phosphoinositide-3-kinase (PI3K)/Akt signaling pathway, Akt, sometimes referred to as protein kinase B, is essential for cellular homeostasis and neuronal survival. The active conformation of Akt and its functional involvement in intracellular signaling are shown by structural models like PDB ID: 1UNQ [13]. One of the most crucial survival mechanisms in the nervous system, activation of the PI3K/Akt pathway in neurons shields cells against apoptosis, oxidative stress, mitochondrial malfunction, excitotoxicity, and neuroinflammation [14,15].

Growth factors such as brain-derived neurotrophic factor (BDNF), nerve growth factor (NGF), insulin-like growth factor-1 (IGF-1), or insulin bind to their specific receptors, activating PI3K and converting PIP2 to PIP3. When PIP3 builds up, Akt is drawn to the plasma membrane, where it is phosphorylated and fully activated [16]. Suppression of pro-apoptotic proteins, increase of antioxidant defense, suppression of glycogen synthase kinase-3β (GSK-3β), and stimulation of mTOR signaling involved in protein synthesis and neural repair are just a few of the downstream processes that are regulated by activated Akt. Neurodegenerative conditions including Parkinson's disease, Alzheimer's disease, and amyotrophic lateral sclerosis have been connected to dysregulation of PI3K/Akt signaling. The inactive Akt1 kinase domain (PDB ID: 4URO) and other structural investigations provide light on the pathogenic pathways linked to Alzheimer's disease and how defective Akt signaling relates to insulin resistance in the brain [17,18].

2. Materials and Methods

2.1. Retrieval and preparation of ligands

Three-dimensional (3D) structures of selected compounds were downloaded from pubchem in SDF format. The ligand molecules were prepared, for docking by open babel in AutoDock. The roots were detected and torsion count was done. The prepared molecules were saved in PDBQT format

2.2. Retrieval and preparation of Protein.

1UNQ and 4URO was chosen as the target protein for this study due to its significant role in the progression of diabetes. The three-dimensional structure was obtained in PDB format from the Protein Data Bank (Figure 1). To prepare the protein for docking studies, unnecessary components such as water molecules, heteroatoms were removed and further processing was done using AutoDock tools, where polar hydrogens were added Kollman charges (−4.666) were assigned. A three-dimensional grid box was defined to blind docking, with dimensions of 126× 126 × 126 Å, ensuring the target region was accurately covered. After all modifications and energy minimization, the final prepared protein structure was saved in PDBQT format, making it ready for molecular docking simulations [19,20].

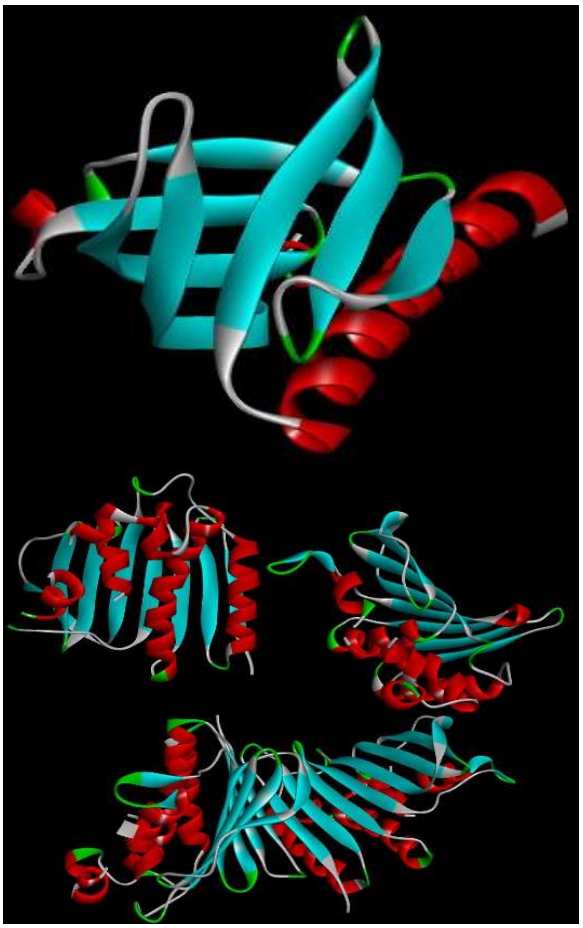


Fig 1: (a) 1UNQ and (b) 4URO 3D structures utilized in the study.

2.3. Binding affinity and molecular docking

The AutoDock program was used to execute blind docking using the target protein and ligand molecules produced PDBQT files. An out.pdbqt file with details on different binding conformations was created from the docking data. Protein–ligand complexes were created using these output files and the protein structure; they were then stored in PDB format for analysis. BIOVIA Discovery Studio (3D) was used to see and study the interactions of the docked complexes. The complexes with the strongest hydrogen bond interactions and the lowest binding energies were selected for more study, suggesting that they might be effective treatment options [21].

2.4. Preparation of stock

For the cytotoxicity tests, a stock solution of the extract was made by dissolving the crude material in DMSO and passing it through a 0.22 µm membrane filter to ensure sterility. Fresh working concentrations were made in the whole culture medium [22].

2.5. Justification for Selection of Test system

- **SHSY5Y cells** are derived from mouse embryonic fibroblasts and are widely used as a model system for studying neurodegenerative diseases, adipogenesis, obesity, diabetes, and lipid metabolism. These cells are capable of differentiating into adipocyte-like cells under appropriate conditions, making them a valuable tool for investigating metabolic regulation and the effects of bioactive compounds. SHSY5Y cells are also relatively easy to culture and maintain in vitro, which makes them suitable for cytotoxicity screening using MTT assays [23].
- Since the MTT assay relies on the reduction of MTT to formazan by mitochondrial dehydrogenases, cell types with active mitochondria provide more reliable and quantifiable results. SHSY5Y cells possess functional mitochondria and exhibit moderate mitochondrial activity both in their preadipocyte and differentiated adipocyte states, making them appropriate for assessing cell viability, proliferation, and metabolic activity using MTT-based assays [24].

2.6. Solubility

The highest concentration (10mg/mL) of TI was dissolved and vortexed for proper mixing [25].

2.7. Selection of Dose

The neuroprotection was determined up to the highest concentration 10 mg/ml [26].

2.8. Formulation

The testing molecule was dissolved and filtered using 0.22 µm filter for sterilization.

Serial two-fold dilutions will be prepared in the selected broth medium to generate a concentration range, 1 mg/ml, 3 mg/ml, 5 mg/ml, 7 mg/ml and 10 mg/ml [27].

2.9. Study Design

For evaluation of cytoprotective potential MTT Assay was performed, and absorbance was measured at 570 nm using a microplate reader.

Neuroprotective Assay	
MTT Concentration	10mg/mL in PBS
Seeding Density	1x 10^4 cell per well
Absorbance	570 nm under microplate reader

2.10. MTT Assay

- For preparation of MTT solution, MTT was added into PBS to make it 5mg/mL
- MTT solution was sterilized through 0.2 µM filter into a sterile, light protected container.
- The MTT solution was stored at 4°C for frequent use or at -20°C for long term storage. For assay protocol 10000 cells per well were seeded in 96-well plates containing a final volume of

100 µL/well in Triplicates along with vehicle and positive control.

- Cells was incubated for 24-48 hours.
- Then MTT solution was added to cells and incubate the plates in Incubator for 4h at 37°C.
- 100 µL solubilization solutions was added to each well to dissolve formazan crystals and ensure the complete Solubilization.
- After 20-30 minutes of adding Solubilization solution absorbance was recorded at 570 nm [28,29].

2.11. Observation

- For Cytotoxicity assay absorbance value was measured at 570 nm. Increase in absorbance indicates higher cell viability and lower toxic effect. IC_{50} (Half-Maximal Inhibitory Concentration) was also measured to estimate the cytotoxicity.

$$\% \text{ Inhibition} = \frac{1 - \text{Mean Absorbance of treated cells}}{\text{Mean Absorbance of control}} \times 100$$

- IC_{50} results was determined from the linear regression graph, % inhibition versus test concentrations [30,31].

3. Result

3.1. Binding affinity and interactions in molecular docking

A range of interaction strengths among the chosen phytochemicals was shown by the docked protein–ligand complexes' binding affinities, which varied from -7.6 to -6.4 kcal/mol. With the lowest binding energy, the ligands indicated a substantial potential interaction. Nevertheless, in spite of this positive score, the complex showed no hydrogen bonds, suggesting that there were no substantial polar interactions between the protein targets. On the other hand, the majority of the ligands interacted with the protein by alkyl or Van der Waals forces. These non-covalent interactions imply that the substances may bind to both the active site and allosteric areas, which might indirectly modulate the protein's activity. This demonstrates the ligands varied binding patterns and their capacity to alter protein activity via a variety of binding mechanisms.

I. 1UNQ docking interactions

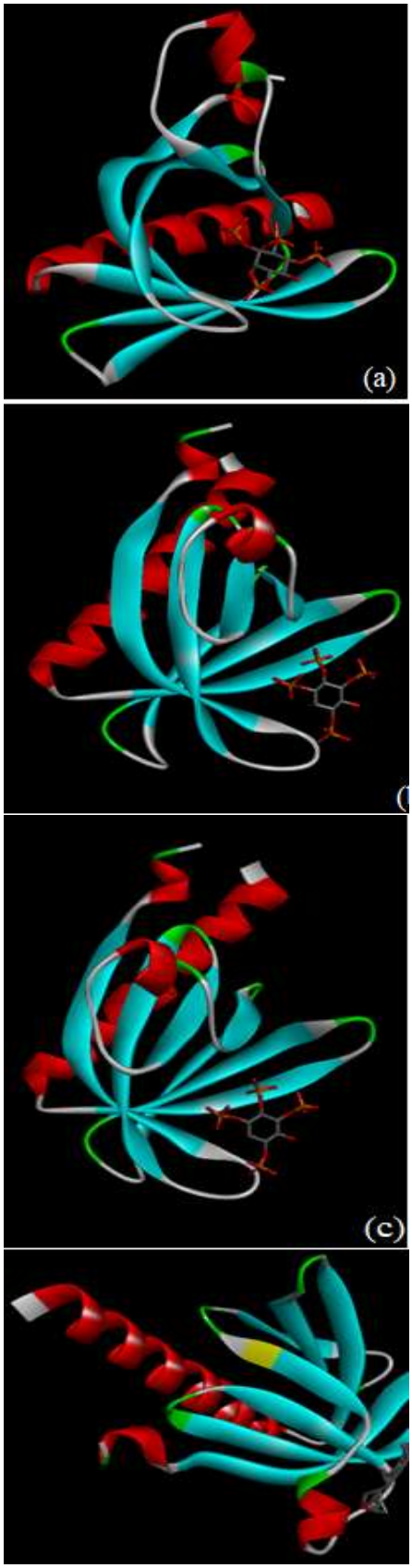


Fig 2 (a-d): Illustrates the 3D interaction between the 1UNQ and the ligands.

II. 4URO docking interactions

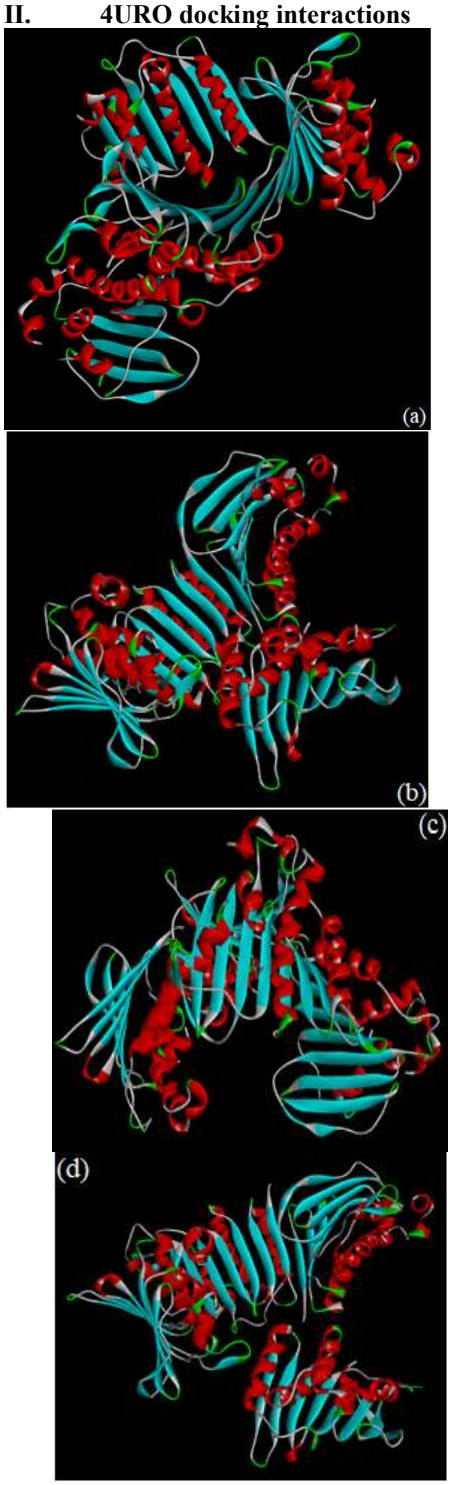


Fig 2 (a-d): Illustrates the 3D interaction between the 4UNQ and the ligands.

S.	Ligands	(IUNQ)	Lowest binding energy in negative	(4URO)	Lowest binding energy in negative
	Diallyl sulfide	15	4.13	88	4.04
	alpha pinene	4	5.07	53	5.11
	Beta pinene	66	4.72	15	4.76
	p-cymene	69	5.44	97	5.21
	Raspberry Ketone	60	5.37	54	5.02

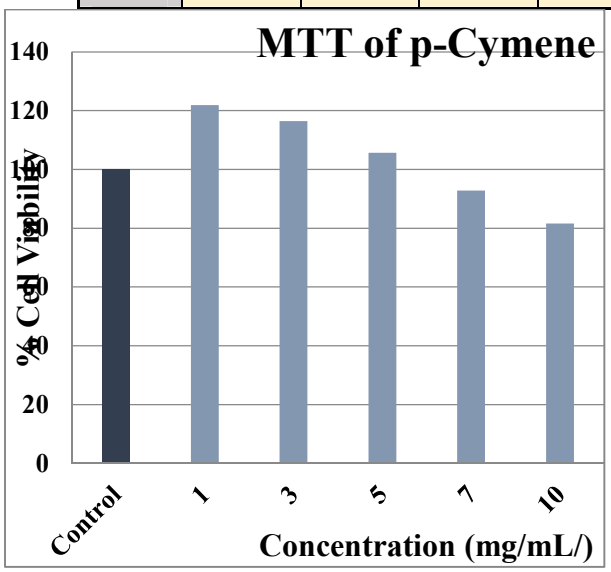
3.2. Cell line studies

The MTT assay results demonstrated that p-Cymene maintains cell viability above the cytotoxic threshold across all tested concentrations. According to established cytotoxicity criteria (≥80% viability), all concentrations evaluated can be considered non-cytotoxic. Notably, lower concentrations (1 mg/mL and 3 mg/mL) significantly enhanced cell viability to 121.89% and 116.46%, respectively, indicating a stimulatory effect on cellular metabolism and suggesting potential neuroprotective activity. At 5 mg/mL, viability remained above baseline (105.65%), further supporting its safety and functional compatibility with cells.

Although a gradual decline in viability was observed at higher concentrations (92.86% at 7 mg/mL and 81.60% at 10 mg/mL), the values remained within the non-toxic range. However, this reduction indicates the onset of cellular stress at elevated doses. Overall, these findings suggest that p-Cymene is non-cytotoxic across the tested range, with optimal neuroprotective potential observed at lower concentrations (1–5 mg/mL), where enhanced cell viability reflects improved cellular resilience and metabolic activity.

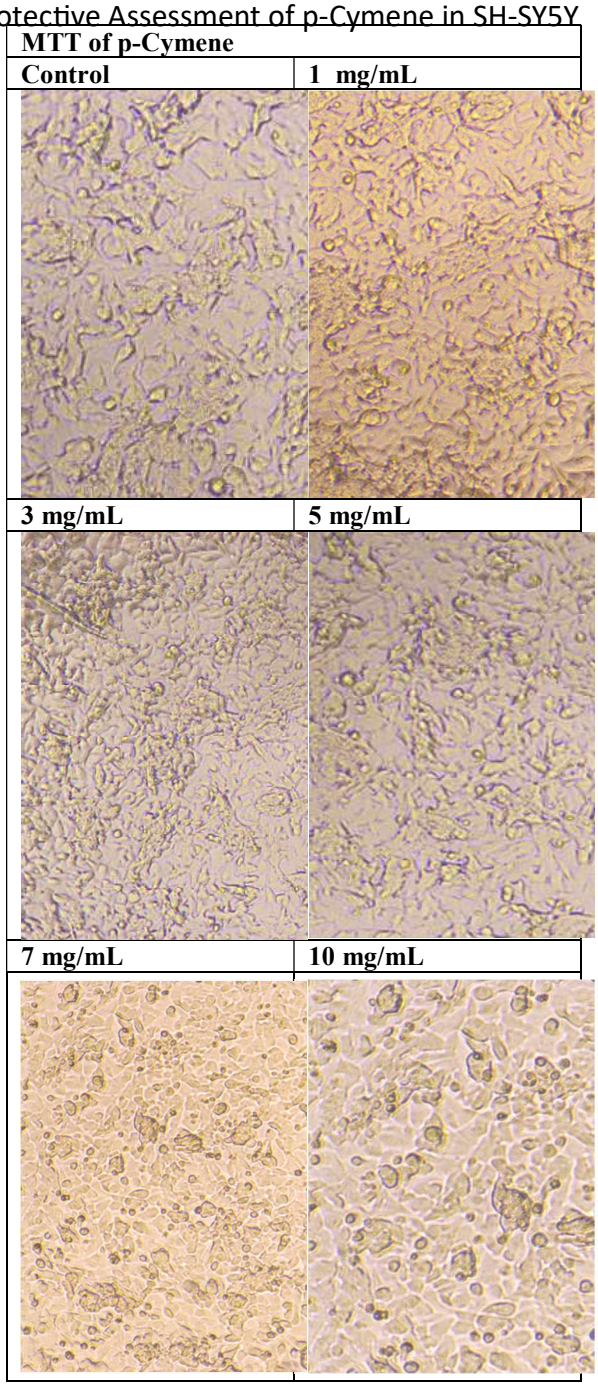
MTT of p-Cymene				
Sample Group	**Replicate 1**	**Replicate 2**	**Replicate 3**	**Mean**
Control	100.0000	100.0000	100.0000	100.0000

1 mg/mL	123.1634	119.0888	123.4226	121.8916
3 mg/mL	115.4217	118.4961	115.4587	116.4588
5 mg/mL	101.6792	110.3099	104.9759	105.6550
7 mg/mL	92.3818	95.5674	90.6408	92.8633
10 mg/mL	81.5286	81.4545	81.8182	81.6004



With increasing concentrations of p-Cymene, mild and gradual morphological alterations were observed in the treated cells, consistent with a dose-dependent response. In the control group, cells exhibited a healthy, dense monolayer with a typical elongated morphology. At 1 mg/mL, cells appeared predominantly normal with intact morphology and no noticeable signs of damage, supporting enhanced cell viability. Similarly, at 3 mg/mL, the majority of cells retained their structural integrity, with only minimal and early signs of cellular stress, such as slight rounding or minor discontinuities in the monolayer.

At 5 mg/mL, a modest reduction in cell density and occasional cell detachment were observed; however, the overall monolayer structure remained largely preserved, indicating maintenance of cellular integrity. At higher concentrations (7 mg/mL and 10 mg/mL), more evident morphological changes such as increased cell rounding, partial detachment, and mild loss of confluence were noted. Despite these alterations, the overall cell population remained largely intact, correlating with viability values remaining above the non-cytotoxic threshold (≥80%).



Overall, the morphological observations suggest that p-Cymene does not induce severe cytotoxic damage within the tested concentration range. Lower concentrations (1–5 mg/mL) maintain normal cellular architecture and may support cellular health, while higher concentrations (7–10 mg/mL) show early signs of cellular stress without extensive structural damage, supporting its potential safety and neuroprotective profile within an optimal concentration range.

3.3. IC50 Studies

The MTT assay results demonstrated a concentration-dependent biphasic response of p-cymene in SH-SY5Y cells. Lower concentrations (1–5 mg/mL)

enhanced cell viability above the control, indicating potential neuroprotective activity through improved mitochondrial function and cellular metabolism. In contrast, higher concentrations (7–10 mg/mL) showed a gradual decline in viability, reflecting the onset of mild cytotoxic stress. However, since cell viability remained above 80% at all tested concentrations, p-cymene can be considered non-cytotoxic within the investigated dose range. These findings suggest that p-cymene possesses promising neuroprotective properties at lower doses while exhibiting minimal cytotoxicity at elevated concentrations.

Concentration (mg/mL)	**Mean Cell Viability (%)**	**% Inhibition**	**Interpretation of Neuroprotective/ Cytotoxic Effect**
Control	100.00	0.00	Represents normal healthy SH-SY5Y cells without treatment.
1 mg/mL	121.89	-21.89	Significant increase in cell viability indicates strong neuroprotective and proliferative activity of p-cymene. Enhanced mitochondrial activity and improved cellular metabolism may contribute to this effect.
3 mg/mL	116.46	-16.46	Cells maintained excellent viability with mild stimulatory effects, suggesting continued neuroprotective potential and absence of cytotoxicity.
5 mg/mL	105.66	-5.66	Cell viability remained above the control level, demonstrating good cellular compatibility and non-cytotoxic behavior.
7 mg/mL	92.86	7.14	Slight reduction in viability suggests the onset of mild cellular stress; however, viability remained above the accepted non-cytotoxic threshold (≥80%), indicating relative safety.
10 mg/mL	81.60	18.40	Moderate decline in cell viability indicates early dose-dependent cytotoxic effects. Despite reduced viability, cells remained within the non-cytotoxic range, suggesting limited toxicity at higher concentrations.

Conclusion

The present study provides a comprehensive evaluation of the cytotoxic profile of *p*-cymene through an integrated *in silico* and *in vitro* approach. Molecular docking analysis revealed favorable binding affinities between selected phytochemicals and the target protein, primarily mediated by non-covalent interactions such as van der Waals and alkyl forces. These interactions suggest a potential ability of monoterpenes to modulate protein function, supporting their pharmacological relevance and justifying further biological evaluation.

The *in vitro* cytotoxicity assessment using the MTT assay in SH-SY5Y cells demonstrated a clear concentration-dependent cellular response to *p*-cymene. At lower concentrations (1–3 mg/mL), *p*-cymene did not induce toxicity and instead showed enhanced cell viability compared to control, indicating possible stimulation of mitochondrial activity or adaptive cellular mechanisms. This observation highlights the relative safety of low-dose exposure and suggests that *p*-cymene may exert protective or metabolic modulatory effects under controlled conditions.

However, as the concentration increased, a gradual reduction in cell viability was observed, with pronounced cytotoxic effects at higher doses (7–10 mg/mL). Morphological changes such as cell rounding, detachment, fragmentation, and loss of confluence further confirmed dose-dependent cellular stress and damage. These findings emphasize the biphasic nature of *p*-cymene, where beneficial or neutral effects at low concentrations transition into cytotoxic outcomes at elevated doses.

Overall, the results underscore the importance of dose optimization when considering *p*-cymene for therapeutic or pharmaceutical applications. While the compound exhibits promising bioactivity and acceptable safety at lower concentrations, its cytotoxic potential at higher doses warrants careful evaluation. Further mechanistic studies, along with *in vivo* investigations, are essential to better define its therapeutic window and clinical relevance.

Reference

1. Schippmann U, Leaman DJ, Cunningham AB. Biodiversity and the ecosystem approach in agriculture, forestry and fisheries. In: Satellite event on the occasion of the ninth regular session of the Commission on Genetic Resources for Food and Agriculture. Rome: FAO; 2002. p. 12–13.
2. Karpiński TM, Adamczak A. Anticancer activity of bacterial proteins and peptides. Pharmaceutics. 2018;10(2):54. doi:10.3390/pharmaceutics10020054.
3. Sharma V, Sarkar IN. Bioinformatics opportunities for identification and study of medicinal plants. Brief Bioinform. 2013;14(2):238–250. doi:10.1093/bib/bbs021.
4. Ranjan A, Ramachandran S, Gupta N, Kaushik I, Wright S, Srivastava S, et al. Role of phytochemicals in cancer prevention. Int J Mol Sci. 2019;20(20):4981. doi:10.3390/ijms20204981.
5. Patridge E, Gareiss P, Kinch MS, Hoyer D. An analysis of FDA-approved drugs: natural products and their derivatives. Drug Discov Today. 2016;21(2):204–207. doi:10.1016/j.drudis.2015.01.009.
6. Mostafa M, Nahar N, Mosihuzzaman M, Makhmoor T, Choudhary MI, Rahman AU. Free radical scavenging phenylethanoid glycosides from Leucas indica Linn. Nat Prod Res. 2007;21(4):354–361. doi:10.1080/14786410701194401.
7. Gogoi D, Arora N, Kalita B, Sarma R, Islam T, Ghosh SS, et al. Anticoagulant mechanism, pharmacological activity, and assessment of preclinical safety of a novel fibrin(ogen)olytic serine protease from leaves of Leucas indica. Sci Rep. 2018;8:6210. doi:10.1038/s41598-018-24422-y.
8. Divya TM, Soorya VC, Amithamol KK, Juliet S, Ravindran R, Nair SN, et al. Acaricidal activity of alkaloid fractions of Leucas indica Spreng against Rhipicephalus (Boophilus) annulatus tick. Trop Biomed. 2014;31(1):46–53.
9. Ramalingam R, Bindu KH, Madhavi BB, Nath AR, David B. Pharmacognostical, phytochemical and anthelmintic evaluation of Leucas indica (L). Phcog J. 2010;2(12):317–323. doi:10.1016/S0975-3575(10)80102-6.
10. Ramani R, Sudini S, Boddupalli BM, Anisetti RN. Antioxidant, free radical scavenging and in vitro cytotoxic studies of ethanolic extract of Leucas indica var lavandulifolia and Leucas indica var nagalapuramiana. Asian Pac J Trop Biomed. 2012;2:S1637–S1642. doi:10.1016/S2221-1691(12)60468-7.
11. Mohan A, Nair SV, Lakshmanan VK. Leucas aspera nanomedicine shows superior toxicity and cell migration retarded in prostate cancer cells. Appl Biochem Biotechnol. 2017;181(4):1388–1400. doi:10.1007/s12010-016-2291-5.
12. Chavata R, Datchanamurthy S, Kotteeswaran V. Biofabrication of silver nanoparticles from aqueous leaf extract of Leucas aspera and their anticancer activity on human cervical cancer cells. Adv Nat Sci Nanosci Nanotechnol. 2019;10(4):045008. doi:10.1088/2043-6254/ab5103.
13. Lourenço SC, Moldão-Martins M, Alves VD. Antioxidants of natural plant origins: from sources to food industry applications. Molecules. 2019;24(22):4132. doi:10.3390/molecules24224132.
14. Xu DP, Li Y, Meng X, Zhou T, Zhou Y, Zheng J, et al. Natural antioxidants in foods and medicinal plants: extraction, assessment and resources. Int J Mol Sci. 2017;18(1):96. doi:10.3390/ijms18010096.
15. Li AN, Li S, Zhang YJ, Xu XR, Chen YM, Li HB. Resources and biological activities of natural polyphenols. Nutrients. 2014;6(12):6020–6047. doi:10.3390/nu6126020.
16. Ogbole OO, Segun PA, Adeniji AJ. In vitro cytotoxic activity of medicinal plants from Nigeria ethnomedicine on Rhabdomyosarcoma cancer cell line and HPLC analysis of active extracts. BMC Complement Altern Med. 2017;17:1–10. doi:10.1186/s12906-017-2005-8.
17. Fang J, Liu C, Wang Q, Lin P, Cheng F. In silico polypharmacology of natural products. Brief Bioinform. 2018;19(6):1153–1171. doi:10.1093/bib/bbx045.
18. Sinan KI, Dall'Acqua S, Ferrarese I, Mollica A, Stefanucci A, Glamočlija J, et al. LC-MS

based analysis and biological properties of Pseudocedrela kotschyi (Schweinf.) Harms extracts: a valuable source of antioxidant, antifungal, and antibacterial compounds. Antioxidants (Basel). 2021;10(10):1570. doi:10.3390/antiox10101570.

19. Mahmud S, Uddin MAR, Paul GK, Shimu MSS, Islam S, Rahman E, et al. Virtual screening and molecular dynamics simulation study of plant-derived compounds to identify potential inhibitors of main protease from SARS-CoV-2. Brief Bioinform. 2021;22(2):1402–1414. doi:10.1093/bib/bbaa428.
20. Hu M, Chen X, Ma L, Ma Y, Li Y, Song H, et al. AMPK inhibition suppresses the malignant phenotype of pancreatic cancer cells in part by attenuating aerobic glycolysis. J Cancer. 2019;10(8):1870–1878. doi:10.7150/jca.28299.
21. Otto T, Sicinski P. Cell cycle proteins as promising targets in cancer therapy. Nat Rev Cancer. 2017;17(2):93–115. doi:10.1038/nrc.2016.138.
22. García-Reyes B, Kretz AL, Ruff JP, von Karstedt S, Hillenbrand A, Knippschild U, et al. The emerging role of cyclin-dependent kinases (CDKs) in pancreatic ductal adenocarcinoma. Int J Mol Sci. 2018;19(10):3219. doi:10.3390/ijms19103219.
23. Roghini R, Vijayalakshmi K. Phytochemical screening, quantitative analysis of flavonoids and minerals in ethanolic extract of Citrus paradisi. Int J Pharm Sci Res. 2018;9(11):4859–4864. doi:10.13040/IJPSR.0975-8232.9(11).4859-64.
24. De S, Dey YN, Ghosh AK. Phytochemical investigation and chromatographic evaluation of the different extracts of tuber of Amorphophallus paeoniifolius (Araceae). Int J Pharm Biomed Res. 2010;1(5):150–157.
25. Joshi RK. GC/MS analysis of the essential oil of Leucas indica from India. Nat Prod Commun. 2014;9(11):1607–1608. doi:10.1177/1934578X1400901119.
26. Shukla PK, Srivastava A, Misra A, Srivastava S. Simultaneous quantification of quercetin and syringic acid in methanolic extract of Leucas lavandulifolia by using validated HPTLC-densitometric method. Indian J Pharm Educ Res. 2020;54(3 Suppl):S687–S694. doi:10.5530/ijper.54.3s.169.
27. Hadni H. 2D and 3D-QSAR, molecular docking and ADMET properties in silico studies of azaaurones as antimalarial agents. New J Chem. 2020;44(16):6553–6565. doi:10.1039/C9NJ05767F.
28. Ghasemi M, Turnbull T, Sebastian S, Kempson I. The MTT assay: utility, limitations, pitfalls, and interpretation in bulk and single-cell analysis. Int J Mol Sci. 2021;22(23):12827. doi:10.3390/ijms222312827.
29. Surin AM, Sharipov RR, Krasil'Nikova IA, Boyarkin DP, Lisina OY, Gorbacheva LR, et al. Disruption of functional activity of mitochondria during MTT assay of viability of cultured neurons. Biochemistry (Mosc). 2017;82(6):737–749. doi:10.1134/S0006297917060104.
30. Ismail N, Ab Ghani NB, Rusli SNA, Abdullah NA, Awang RA. Cytotoxicity assessment of zirconia-reinforced experimental nanohybrid dental composite using MTT assay. Ann Rom Soc Cell Biol. 2021;25:14878–14886.
31. Ong THD, Yu N, Meenashisundaram GK, Schaller B, Gupta M. Insight into cytotoxicity of Mg nanocomposites using MTT assay technique. Mater Sci Eng C Mater Biol Appl. 2017;78:647–652. doi:10.1016/j.msec.2017.04.129.