

Anti-Amnesic Effect of Methanolic Leaf Extract of *Tecoma stans*: An Experimental Study in Rodents

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ABSTRACT

Objectives: Herbal therapy is a natural practice of curative or substitute remedy to treat disorders or illnesses aimed at the management of cognitive ailments. Memory loss is a shortfall in recollection triggered by brain impairment or illness, but it can also be initiated briefly by the usage of numerous tranquilizing and hypnotic medications. **Methods:** In present study the methanolic leaf extract of *Tecoma stans* (METS) was screened for its Anti-amnesic activity in rodent models. The various phytochemical constituents like alkaloids, flavonoids, phenolics, terpenoids, tannins, sterols, saponins, glycosides, amino acids and carbohydrates present in the extract were identified by using GC-MS analysis. **Results:** Toxicity studies revealed that the extract was safe upto 2000 mg/kg, bd.wt. as per OECD guidelines 425. *In-vivo* anti-amnesic activity was accomplished in diazepam and aluminium induced amnesic models by actophotometer, rotarod and cook's pole climbing apparatus. Various biochemical estimates like AChE, TBARS, GSH and SOD and histopathological studies of mice brain were performed in aluminium induced amnesic model. The *in-vitro* cholinesterase activity is evaluated using Ellman's assay. METS exhibited noteworthy enhancement

in intellectual diminishing in diazepam and aluminium chloride induced amnesic models and also suggestively decreased brain AChE and oxidant enzyme factors like TBARS, GSH and SOD levels and moderate inhibitory activities in ChE's assay. **Conclusion:** From the results it is clear that the methanolic leaf extract of *Tecoma stans* possess anti-amnesic activity.

Key words: *Tecoma stans*, Anti-amnesia, Memory, Acetylcholinesterase, Aluminium chloride, Ellman's assay.

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INTRODUCTION

Dementia is a parasol stretch that shields diverse kinds of scientific conditions such as Alzheimer's disease (AD), vascular dementia, dementia with Lewy bodies, fronto-temporal dementia, Parkinson's disease, etc. In 2001, 24.3 million individuals had dementia and 4.6 million novel circumstances are diagnosed each time. It has been predicted that the number may double in next 20 years.¹ In future India will have one of the largest number of elderly with dementia. Memory loss is a shortage in memory caused by injury to the brain or illness, but it can also be produced momentarily by the usage of numerous tranquilizer medications. Recollection can be either completely or partly absent owing to the degree of impairment that was affected. At hand are two foremost kinds of oblivion: anterograde amnesia and retrograde amnesia. Anterograde amnesia is more severe than retrograde amnesia. Stroke, infections, dementia, Alzheimer's disease, improper oxygen supply to the brain, alcohol abuse, lack of acetylcholine and certain medications are also the causative agents of amnesia.^{2,3}

Diazepam produces memory loss in brain cells of mice by increasing the GABAergic inhibitory facilitation and/or by causing oxidative stress by releasing free radicals in the brain cavity. GSH plays a vital part in shielding the brain from free radical release by neutralizing these free radicals. The harm by free radicals to brain soft tissue is connected with neurodegenerative ailments. Diazepam can weaken interim reminiscence and knowledge of novel facts and leads to anterograde amnesia, but do not cause retrograde amnesia.⁴

Aluminium (Al) is a permeating lethal metal that chiefly affects the mind, skeleton, liver and spleen, whereas the accretion of Al deposits

in the brain and its successive deadliness leads to neurodegeneration.^{5,6} treatment with AlCl₃ (100 mg/kg) considerably raised the levels of aluminium (Al), acetylcholinesterase (AChE) and amyloid precursor protein (APP), β amyloid (A β 1-42), β and γ secretases, progress of oxidative stress and neuronal apoptosis in the hippocampus which is a place for reminiscence formation and synaptic plasticity occurs during learning which is similar to the pathogenesis of AD.⁷

Tecoma stans is an erect, large flowering perennial shrub, spear shaped green leaves, branched, yellow bright trumpet like flowers and 5-7 meters in height, commonly found in India and belongs to the family *Bignoniaceae*. Leaves of *Tecoma stans* are used to cure diabetes, stomach pains, digestive and diuretic problems and are also reported to possess anti-cancer, wound healing, anti-inflammatory,⁸ anti-spasmodic, analgesic, anti-hyperlipidemic, antifungal, anti-microbial, anti-arthritis, anti-ulcer and anti-oxidant activity.⁹ The main objective of this exploration was to assess the anti-amnesic activity of the methanolic extract of leaves of *Tecoma stans*.

MATERIALS AND METHODS

Chemicals and drugs

Methanol, chloroform, ethyl acetate, acetylcholine iodide, DTNB, disodium hydrogen phosphate and sodium dihydrogen phosphate were purchased from SD Fine Chemicals Limited, Mumbai. Donepezil was purchased from Hetero laboratories, Hyderabad, Diazepam from Ranbaxy laboratories and Aluminium chloride from Himedia laboratories Mumbai.

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Plant collection and authentication

The leaves of *Tecoma stans* were collected near Osmania University, Hyderabad during the month of December and authenticated through a Botanist, Dr. Rabiya Sultana, Osmania University, Hyderabad.

Preparation of extract and fractionation

The desiccated constituents of leaves of *Tecoma stans* (5 kg) were crushed into coarse powder and extracted through soxhlation using methanol at room temperature for 5 days. The crude methanol extract was subjected to partial fractionation with solvents like chloroform, methanol and ethyl acetate. The crude methanolic extract was partitioned with chloroform and water (2:1). From the chloroform and aqueous layer, aqueous layer was again fractionated with methanol and ethyl acetate (8:2) by using separating funnel. The obtained methanol and ethyl acetate fraction was taken and were evaporated to dryness and used further.¹⁰

Preliminary phytochemical analysis

The extract was subjected to primary phytochemical investigations to identify various constituents present in the leaves of *Tecoma stans*.¹¹

Experimental Protocol

Animal procurement

Swiss albino mice (approx. 20 to 25 g) were procured from Albino research Laboratories, Hyderabad. The study was reviewed and permitted by the IAEC, (Reg. No. 1175/PO/Re/S/08/CPCSEA), GRCP, Bachupally, Hyderabad, India.

Animal housing

The animals were accommodated in poly acrylic cages with six animals per cage, with light-dark cycle. Mice are allowed to have standard diet and drinking water. The mice were permitted to adjust the workroom atmosphere for a week earlier to the trial. The caution and preservation of the animals were agreed as per accepted procedures of the CPCSEA.

In-vivo methods for evaluation of Anti-amnesic activity

A. Diazepam induced (acute) amnesic model

The *in vivo* evaluation of anti-amnesic activity of the METS leaves was evaluated in the diazepam induced (acute) amnesic model using Actophotometer, Rotarod and Cook's pole climbing apparatus. At predetermined time intervals, i.e. on the 8th and 9th day the interactive constraints like basal activity score, fall off time and passive avoidance time were evaluated and the design of the study were given in the Table 1, 2 and 3.¹² 30 Healthy Swiss albino mice of both sex weighing 20-25 g were carefully chosen and these were divided into 5 groups ($n=6$) in each group.

B. Aluminium chloride induced (chronic) amnesic model

The *in vivo* evaluation of anti-amnesic activity of the methanolic extract of *Tecoma stans* was evaluated in Aluminium chloride induced (chronic) amnesic model using Cook's pole climbing apparatus and Elevated plus Maze.¹³ At the predetermined time intervals, i.e. on 20th, 21st and 42nd day the behavioral parameters like a transfer latency time using elevated plus maze and time taken to climb pole using the cook's pole climbing apparatus were evaluated and study design is depicted in Table 4 and 5.

30 Healthy Swiss albino mice of both sex weighing 20-25 g were carefully chosen and these were divided into 5 groups ($n=6$ in each group).

Biochemical estimations

At the finish of the protocol mice were forfeited by cervical disarticulation and brains were carefully detached and normalized in phosphate buffer pH 7.4 and centrifuged at 4500 rpm for 15 min and supernatants were

Table 1: Effect of METS on basal activity score using Actophotometer.

Groups	Treatment	Basal Activity Score	
		8 th day	9 th day
I	Control	324.16 ± 0.87	320 ± 0.63
II	Disease control (Diazepam 1mg/kg)	185.33 ± 0.76**A	167.16±0.74**A
III	METS (200 mg/kg)	196.16 ± 0.47**aA	246 ± 0.57**aA
IV	METS (400 mg/kg)	201.83 ± 0.70**aA	300.66 ± 0.66**aA
V	Donepezil (1 mg/kg)	210.66 ± 0.49**a	330.33 ± 0.76**a

Table 2: Effect of METS on fall off time using rotarod.

Groups	Treatment	Fall off time (secs)	
		8 th day	9 th day
I	Control	249 ± 0.57	244.16 ± 0.47
II	Disease control (Diazepam 1mg/kg)	98.16 ± 0.74**A	76.33 ± 0.49**aA
III	METS (200 mg/kg)	118.83 ± 0.47**aA	175.5 ± 0.42**aA
IV	METS (400 mg/kg)	143 ± 0.36**aA	218.33 ± 0.57**aA
V	Donepezil (1 mg/kg)	150.83 ± 0.83**a	250 ± 0.57**a

Values are stated as Mean ± SEM, ($n=6$). All the groups were related with control, disease control and standard using Dunnett's test. Noteworthy values were expressed as control group (** $p<0.01$), disease control (a= $p<0.01$) and standard (A= $p<0.01$).

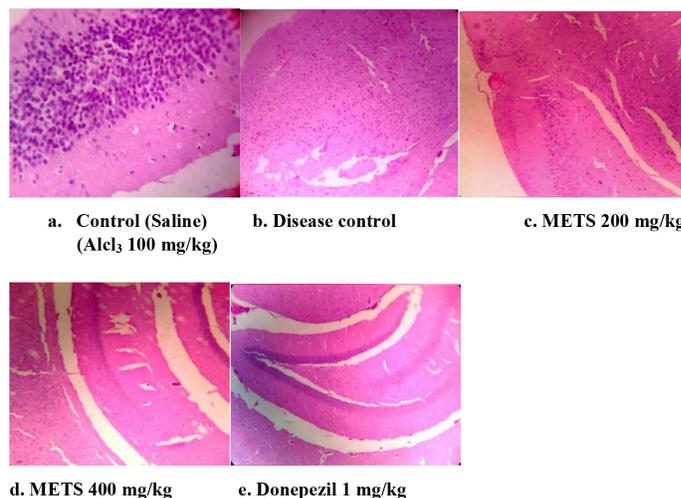


Figure 1: Histopathological images of groups treated with saline, aluminium chloride, METS and standard drug Donepezil.

collected and used for the estimation of AChE activity, TBARS, GSH and SOD.¹⁴⁻¹⁷

Histopathological Studies

In the $AlCl_3$ model after 42 days, one mouse each from five groups was euthanized and the brains were isolated *via* clearing its extraneous connections with cranium. Later it was cut in to two vertical halves. Hippocampus was separated from the rest of the brain and stored in 10% formalin solution; these specimens were stored and used for Histopathological studies.

In vitro Evaluation of AChE Inhibitory Activity using Elman's Inhibition Assay

The acetyl cholinesterase inhibitory activity of the plant extract was evaluated by the Ellman reagent. Concisely, 0.1 M Na₃PO₄ buffer (pH 8.0, 150 µL), 10 µL of test *i.e.* extract at the concentrations (10-50 µg/mL) and enzyme solution (brain homogenate of mice) (0.1 units/mL, 20 µL) were assayed and reared for 15 min at 25°C. 10 µL of DTNB (10 mM) was added and response was started by the adding the substrate (10 µL of Acetyl thiocholine iodide, 14 mM solution). Acetyl thiocholine iodide hydrolysis can be estimated through the creation of the coloured product 5-thio-2-nitrobenzoate anion formed by the reaction of DTNB (5, 5-dithio-bis-[2-nitro benzoic acid] and thiocholine, which is free via the cleavage of enzyme. The creation of the tinted produce was determined at 410 nm wave length after 10 min. In the assay Donepezil at concentrations of 10 µM was used as a standard with the same procedure as for the test extract. AChE % inhibition was estimated using the formula.¹⁸

$$\text{Inhibition activity (\%)} = \frac{(1 - \text{Absorbance of sample})}{\text{Absorbance of control}} \times 100.$$

Statistical Analysis

Values are stated as Mean ± SEM, (n=6). All the groups were related with control, disease control and standard using Dunnett's test. Noteworthy values were expressed as $p < 0.001$ and $p < 0.05$.

RESULTS

Extraction yield

The methanolic extract of leaves of *Tecoma stans* was prepared via Soxhlation technique. The percentage yield of the extract was found to be 24.54% and the fraction yield is found to be 33.3%.

Preliminary phytochemical analysis

The preliminary study of the methanolic extract of leaves of *Tecoma stans* exhibited the occurrence of alkaloids, flavonoids, phenolics, sterols, terpenoids, tannins, saponins, glycosides, amino acids and carbohydrates.

Acute toxicity studies

Methanolic leaf extract of *Tecoma stans* was tried on Swiss albino mice up to a dose of 2000 mg/kg bd.wt. The animal did not reveal any marks of noxiousness or transience upto 2000 mg/kg bd.wt. Various morphological and behavioural characters were observed during the study. The other parameters like food and water consumption was also observed. All the animals were found to be harmless even after 14 days of surveillance.

Pharmacological evaluations were conducted at 200 and 400 mg/kg, bd.wt. *p.o.*

In vivo Methods for Evaluation of Anti-Amnesic Activity

Anti-amnesic activity of METS was screened against various models like

1. Diazepam induced acute amnesic model.
2. Aluminium chloride induced chronic amnesic model.

In diazepam induced amnesia, the behavioral parameters like basal activity score (BAS), fall off time and passive avoidance time was analyzed by actophotometer, rotarod and cook's pole climbing apparatus. The results were depicted in Table 1, 2 and 3.

In the aluminium induced amnesia the behavioral parameters like the transfer latency time and passive avoidance time was analyzed using elevated plus maze and cook's pole climbing apparatus. These parameters were depicted in Table 4 and 5. Apart from the above-stated behavioral parameters various biochemical parameters like acetylcholinesterase (AChE) using Ellman's assay and Thiobarbituric acid reactive species (TBARS), GSH and SOD levels were also estimated in aluminium chloride induced amnesic model and depicted in Table 6.

1. Diazepam Induced Acute Amnesic Model

- a. Basal activity score using Actophotometer
- b. Fall off time by Rotarod test
- c. Passive avoidance time by Cook's pole climbing apparatus

2. Aluminium Chloride Induced (Chronic) Amnesic Model

- a. Transfer latency time by Elevated plus Maze
- b. Passive avoidance time by Cook's pole climbing apparatus

Table 3: Effect of METS on Passive avoidance time using cook's pole climbing apparatus.

Groups	Treatment	Time taken to climb the pole (secs)	
		8 th day	9 th day
I	Control	12.66 ± 1.27	12.33 ± 0.61
II	Disease control (Diazepam 1mg/kg)	23 ± 0.68	27.16 ± 0.47
III	METS (200 mg/kg)	21.5 ± 0.42**B	18 ± 0.36**aA
IV	METS (400 mg/kg)	19.83 ± 0.74*bB	15.33 ± 0.66*aA
V	Donepezil (1 mg/kg)	16.83 ± 0.57*a	11 ± 0.51a

Table 4: Effect of METS on transfer latency time using elevated plus maze.

Groups	Treatment	Transfer latency time (secs)		
		20 th day	21 st day	42 nd day
I	Control	24.83 ± 0.47	24 ± 0.36	22 ± 0.57
II	Disease control (Diazepam 1mg/kg) (AlCl ₃ 100 mg/kg)	42 ± 0.68**A	45.16 ± 0.60**A	57.33 ± 1.03**A
III	METS (200 mg/kg)	35 ± 0.57**aA	33 ± 0.57**aA	30.66 ± 0.6**aA
IV	METS (400 mg/kg)	33.16 ± 0.7**aB	31.5 ± 0.42**aB	27 ± 0.73**aA
V	Donepezil (1mg/kg)	30 ± 0.96**a	28.5 ± 0.85**a	23.16 ± 0.47a

Values are stated as Mean ± SEM, (n=6). All the groups were related with control, disease control and standard using Dunnett's test. Noteworthy values were expressed as control group (** $p < 0.01$), disease control (a= $p < 0.01$) and standard (A= $p < 0.01$, B= $p < 0.05$).

Table 5: Effect of METS on passive avoidance time using cook's pole climbing apparatus.

Groups	Treatment	Time taken to climb the pole (secs)		
		20 th day	21 st day	42 nd day
I	Control	13.33 ± 0.42	13 ± 0.49	12.5 ± 0.42
II	Disease control (AlCl ₃ 100 mg/kg)	21 ± 0.85 ^{**A}	23 ± 0.73 ^{**A}	28 ± 0.36 ^{**A}
III	METS (200 mg/kg)	19.83 ± 0.89 ^{**aA}	21.5 ± 0.76 ^{**A}	18.16 ± 0.79 ^{**aA}
IV	METS (400 mg/kg)	17.66 ± 0.42 ^a	19 ± 0.85 ^{bb}	15.33 ± 0.66 ^{aA}
V	Donepezil (1mg/kg)	16.83 ± 0.79 ^a	16.16 ± 0.60 ^a	11.66 ± 0.42 ^a

Values are stated as Mean ± SEM, (n=6). All the groups were related with control, disease control and standard using Dunnett's test. Noteworthy values were expressed as control group (**p<0.01, *p<0.05), disease control (a=p<0.01) and standard (A=p<0.01, B=p<0.05).

Table 6: Effect of METS on AChE activity and oxidative stress parameters in brain in aluminium chloride model.

Groups	AChE activity (nM/min/mg of protein)	TBARS (nM/mg of protein)	GSH (nM/mg of protein)	SOD (units/mg of protein)
Control	0.59 ± 0.005	7.6 ± 0.95	46.33 ± 0.84	8.6 ± 0.34
Disease control (AlCl ₃ 100 mg/kg)	1.21 ± 0.010 ^{**A}	18 ± 0.68 ^{**A}	23 ± 1.03 ^{**A}	5.16 ± 0.66 ^{**B}
METS (200 mg/kg)	0.90 ± 0.03 ^{**aA}	13.83 ± 0.98 ^{**aA}	36.83 ± 0.65 ^{**aA}	6.0 ± 0.42 [*]
METS (400 mg/kg)	0.80 ± 0.012 ^{**aA}	12.5 ± 0.71 ^{**a}	38.5 ± 0.42 ^{**aA}	7.3 ± 0.55 [*]
Donepezil (1 mg/kg)	0.71 ± 0.011 ^{**a}	10.66 ± 0.66 ^{**a}	42.33 ± 0.57 ^{**a}	8 ± 0.85 ^b

Values are stated as Mean ± SEM, (n=6). All the groups were related with control, disease control and standard using Dunnett's test. Noteworthy values were expressed as control group (**p<0.01), disease control (a=p<0.01, b=p<0.05) and standard (A=p<0.01, B=p<0.05).

Table 7: AChE inhibition of methanolic extract of leaves of *Tecoma stans*.

S.No	Compounds	Concentrations (µg/ml)	% Inhibition	IC ₅₀ Values
1	METS	10	10.66 ± 0.82	33.2
		20	24 ± 0.81	
		30	46 ± 2.51	
		40	59 ± 2.46	
		50	72.3 ± 0.88	
2	Donepezil	10	15.5 ± 0.89	31
		20	28.5 ± 1.81	
		30	47.1 ± 1.54	
		40	69.6 ± 3.63	
		50	86.5 ± 2.42	

Biochemical Estimations

a. Estimation of whole brain AChE activity

Subsequently behavioral studies the biochemical parameter like AChE activity was estimated in mice brain using Ellman's assay. AlCl₃-treated mice exhibited a substantial rise in brain AChE activity when compared to the control group. Administration of METS (200 and 400 mg/kg, bd.wt, p.o.) and standard drug Donepezil (1 mg/kg, bd.wt, p.o) significantly decreased the AChE levels when compared to disease control.

b. Assessment of whole brain TBARS level

The biochemical parameter like TBARS activity was determined in mice brain AlCl₃ treated mice revealed a major increase in brain TBARS level when compared to the control group. Administration of METS (200 and

400 mg/kg, bd.wt, p.o.) and standard drug donepezil (1 mg/kg, bd.wt, p.o) significantly decreased the TBARS level when compared to disease control.

c. Assessment of whole brain GSH level

The biochemical parameter GSH activity was estimated in mice brain. AlCl₃-treated mice indicated a noteworthy reduction in brain GSH level when compared to the control group. Administration of METS (200 and 400 mg/kg, bd.wt, p.o) and standard drug donepezil (1 mg/kg, bd. wt, p.o) significantly increased the GSH level when compared to disease control.

d. Assessment of whole brain SOD level

The biochemical parameter like SOD activity was estimated in mice brain. AlCl₃ treated mice revealed a substantial decline in brain SOD activity when compared to the control group. Administration of METS (200 and 400 mg/kg, bd.wt, p.o.) and standard drug Donepezil (mg/kg, bd.wt, p.o) significantly increased the SOD when compared to disease control.

In vitro Evaluation of AChE inhibitory activity using Ellman's inhibition assay

In vitro AChE inhibition was performed for methanolic extract of *Tecoma stans* and compared with standard drug donepezil using Ellman's assay and results were given in Table 7.

Subsequently Alzheimer's disease etiopathogenesis is hypothetically accredited to acetylcholine debility and Aβ development; the anti-amnesic properties of METS were estimated via inhibition of AChE. It could be concluded that the alkaloids, flavonoids, phenolics, sterols, terpenoids, tannins, saponins and glycosides from METS might have shown inhibitory activity against AChE. Donepezil is a standard and centrally acting reversible acetyl cholinesterase inhibitor. Donepezil reverses cognitive impairment through inhibition of cholinesterase enzyme.

Histopathology Studies

Histopathology study of hippocampus of the mice in aluminium chloride induced chronic amnesic model. The pathological change was viewed under the light microscope after staining with haematoxylin and eosin were depicted in Figure 1.

Control group: A compactly arranged 7-8 layer of pyramidal cells with the prominent nucleus was observed in hippocampus.

Disease control (AlCl₃, 100 mg/kg, bd.wt, p.o): The absence of pyramidal cells and presence of apoptotic cells was observed in hippocampus.

METS (200 mg/kg, bd.wt, p.o): An Irregular arrangement of 2-3 layers of pyramidal cells with the scattered pattern and mild appearance of apoptotic cells was observed in hippocampus.

METS (400 mg/kg, bd.wt, p.o): Neuronal cells are well organized with 4-5 layers of pyramidal cells were observed in hippocampus.

Donepezil (1 mg/kg, bd.wt, p.o): Neuronal cells are well organized with 6-7 layers of pyramidal cells and the absence of apoptotic cells was observed in hippocampus.

DISCUSSION

In the present study methanolic extract of leaves of *Tecoma stans* was evaluated for anti-amnesic activity using diazepam and aluminium chloride induced amnesic animal replicas and various behavioural parameters like basal activity score, latency to fall off time, passive avoidance time and transfer latency time were evaluated. Apart from the above parameters, the biochemical considerations like AChE, TBARS, GSH and SOD levels are also estimated in brains of mice in the aluminium induced amnesic model.

The various phytochemicals identified in the methanolic extract of leaves of *Tecoma stans* are alkaloids mainly (tecomanine, tecostidine, tecostanine) indolic alkaloids (indole, tryptophan, tryptamine, anthranilic acid, skatole) and other alkaloids (caffeine, boschniakine, 5 β hydroxyl skitanthine, 4-noractinidine) flavonoids (luteolin, quercetin, chrysoeriol, flavones) polyphenols, phenols, sterols, terpenoids, tannins, saponins, iridoid glycosides and many other long chain fatty acids. It can be stated that the above said active constituents like alkaloids, flavonoids, phenols, terpenoids, sterols and many long chain fatty acids have also been reported in GC-MS analytical data which have shown improvement in cognitive function.^{19,20}

Among the phytoconstituents, alkaloids are reflected to be the supreme capable constituents for usage in the treatment of AD, due to their composite nitrogen-containing configurations and also majority of naturally derived inhibitors of AChE are alkaloids. The indole variety of alkaloids is essential natural AChE inhibitors. For indole alkaloids, AChE discernment is supposed to be donated through the existence of alkyl groups as substituents at the indole nitrogen.

Over the last decade, polyphenols have been suggested in the prevention and treatment of cognitive diseases, owing to their anti-oxidative and anti-amyloidogenic features and can decrease A β -induced apoptosis via preventing A β aggregation and/or decreasing ROS. Quercetin, a representative flavonoid increases Brain APoE levels and decreases A β intensities in the cortex of the amyloid model. Luteolin, a flavonoid compound, protects against cognitive dysfunction and reports suggests in a substantial decrease in A β production.²¹

Terpenoids decreases A β creation by controlling the APP method, which is escorted via an enhancement in the perceptive role. Iridoid glycosides improve recall scarcities and diminish hippocampal neuronal damage by refining the brain situation for mending and helping neuronal existence. Omega-3 fatty acids DHA and EPA have also been revealed to progress a perceptive recital in numerous animal studies of AD.^{22,23}

Donepezil acentrally acting reversible acetyl cholinesterase inhibitor is used as a standard. It includes the alterable reticence of cholinesterases (eg. acetylcholinesterase), which inhibits the hydrolysis of acetylcholine and clues to an augmented amount of acetylcholine at cholinergic synapses. Substantiation recommends that the anti-cholinesterase activity of donepezil is comparatively precise for acetylcholinesterase in the brain and henceforth it shows advanced perceptive enactment.²⁴

CONCLUSION

In conclusion, the methanolic extract of *Tecoma stans* has shown the occurrence of bioactive elements like alkaloids, flavonoids, phenolics, sterols, terpenoids, tannins, saponins, glycosides, amino acids and carbohydrates. The animals have shown improvement in cognitive functions which may be owing to the occurrence of active components like alkaloids, flavonoids, phenolics, sterols, terpenoids, tannins and glycosides in the extract. The extract at different dose levels significantly inhibited the cholinesterase enzyme. The assay reports establish a high AChE inhibition activity of the extract. Microscopy studies of brain tissue of mice revealed enhancement of numeral stratum and establishment of pyramidal cells in hippocampus of mice administered with the METS and the donepezil.

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CONFLICT OF INTEREST

All authors have no conflicts of interest to declare.

ABBREVIATIONS

AChE: Acetylcholinesterase; **Al:** Aluminium; **APP:** Amyloid precursor protein; **AD:** Alzheimer's disease; **ATCI:** Acetyl thiocholine iodide; **DTNB:** (5, 5-dithio-bis-[2-nitro benzoic acid]; **TBARS:** Thiobarbituric acid reactive species; **GSH:** Reduced glutathione; **SOD:** Superoxide dismutase; **METS:** Methanolic Extract of *Tecoma stans*; **BAS:** Basal activity score; **APoE:** Apolipoprotein E; **DHA:** Docosahexaenoic acid; **EPA:** Eicosapentaenoic acid; **GC-MS:** Gas chromatography-mass spectrometry

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