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Nootropic activity of Ethanolic extract of *Daucuscarota* Linn. Leaves in mice.

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ABSTRACT

In the present study we have evaluated the Nootropic activity of Ethanolic extract of *Daucuscarota* Linn. Leaves(EDCL)with the various doses (100,250,500mg/kg)in mice. Elevated plus Maze, Morris-Water Maze, Passive avoidance test, Object recognition test, Estimation of Acetyl cholinesterase(AchE) activity was done in mice brain. Piracetam was used as standard drug and Scopolamine was used to produce amnesia in mice. No mice showed mortality up to 5000mg/kg. On, Elevated plus Maze, EDCL shows significantly (*P<0.01*) decrease in Transfer Latency (TL) and increased in inflexion ratio (I.R) compared to vehicle treated group and antagonised effect of scopolamine. On, Morris-Water Maze, Escape Latency significantly (*P<0.01*) decreased by EDCL and piracetam treated groups in mice. On, Passive avoidance test, significant (*P<0.01*) increase in step down latency by EDCL was observed and increased occupancy in the shock free zone (SFZ) of the paradigm and also exhibited diminished preference to the preferred shock zone. The results of Object recognition test, shows EDCL and piracetamantagonises the amnestic effect of scopolamine and it spent more time exploring the novel object. EDCL shows significantly (*P<0.01*) decreased the AChE level in mice brain and hence indicates the involvement of cholinergic system in its mechanism. Thus from the results and observations it is prove that EDCL has potential anti-cholinesterase agent and it enhanced memory retention and protected against scopolamine induced amnesia in mice.

Keywords: Nootropic, *Daucuscarota*Linn. Leaves, Elevated plus maze, Morris-water maze,Acetyl cholinesterase (AchE).

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INTRODUCTION

Memory is ability of an individual to record information, event and retains them over short or long periods of time and recalls them whenever needed. Age, emotion and stress are the conditions that may lead to memory loss, amnesia, dementia, anxiety, and other complications like schizophrenia and Alzheimer's diseases (AD). [1, 2] Dementia is a mental disorder characterized by loss of intellectual ability sufficiently severe as to interfere with occupational or social activities. Dementia is of several types and it invariably involves impairment of memory. The most common cause of dementia is AD, a progressive neurodegenerative disorder associated with loss of neurons in distinct brain area. [3, 4] Central cholinergic pathways plays important role in learning and memory processes. Loss of cholinergic cells particularly in the basal forebrain is accompanied by loss of the neurotransmitter acetylcholine (Ach). A decrease in Achin the brain of patients with AD appears to be a critical element in producing dementia [3, 5]

Nootropics referred to as "smart drugs or memory enhancers", have been shown to improve many aspects of brain and body function along with elevating mood and concentration levels. [6, 7] Nootropics decrease platelet aggregation, increase cerebral blood flow and oxygen consumption, Increase adenylatecyclase breakdown of ADP to ATP and increase density of frontal cortex Ach receptors by 30-40% . [6, 8]

Indian system of medicine suggested use of herbs, nutraceuticals for controlling age related neurodegenerative disorders. Plants like *Ocimum sanctum*, *Tragiaplukenetii*, *Rose alba*, *Azadirachtaindica*, *Withaniasomnifera*, *Celastruspaniculatus* and *Moringaolefera* have been investigated for their nootropic functions. [9-11]

From literature survey the *Daucuscarota* linn. Leaves shows the presence of carotenoids (β-carotene) and *Daucuscarota*linn. Root also contain carotenoids (β-carotene) used in regularly in Indian kitchen. Hence, in the present study we have investigated to study Nootropic activity of *Daucuscarota*linn. Leaves extract.

MATERIALS AND METHODS

Plant material

The fresh leaves of *Daucuscarota* Linn. (Apiaceae) were collected from the local market of Kannad, Aurangabad and plant was authenticated from Botanical Department of Maulana Azad College (Voucher Specimen No.MACH-01243), Dr.Rafiq Zakaria Campus, RouzaBagh, Aurangabad. The collected drug was dried under shade at room temperature for 3 weeks.

Preparation for Extraction of *Daucuscarota*linn. Leaves:

The collected fresh plant materials were dried in shade at room temperature for 3 weeks and they were made in to coarse powder with the use of mortar and pestle. The powder of plant material (500 gm) was extracted with 95 % ethanol(60^0c)using Soxhlet extraction apparatus for 3 days.

Then the extract of ethanol was concentrated. Extract obtained was dried by vaccum evaporator. The yield of ethanolic extract was found to be 16 % w/w. Extract was suspended in distilled water using carboxy methyl cellulose (CMC) 0.2% v/v as the suspending agent prior to administration.

Experimental animals:

Swiss albino mice of either sex weighing between (22-30 g respectively) were used. The animals were allowed to acclimatize to the laboratory conditions for 10 days after their arrival. The animals were housed under standard housing condition. They were maintained at temperature of 25 ± 2°C and relative humidity of 45 to 55% and under standard environmental conditions (12 h. light /12 h. dark cycles).The animals had free access to food and water. All the experiments were carried out between 9 to 18 Hrs. The animals were randomized into experimental and control group and six in groups in sanitized polypropylene cages containing sterile paddy husk as bedding. Animals were habituated to laboratory conditions for 48 hr prior to experimental protocol. The animal were fasted overnight prior to drug administration and during experiment.

The experimental protocol was approved by Institutional Animal Ethics Committee (IAEC) of Y.B. Chavan College of Pharmacy Aurangabad. (CPCSEA/IAEC/ P'col-51/2015-16/114).

Safety Studies

Safety studies were performed according to OECD-423 guidelines category IV substance (acute toxic class method). Swiss albino mice 20-25gm (n=3) of either sex selected by random sampling technique were employed in this study. The animals were fasted for 4 hrs with free access to water. The plant extracts of EDCL were administered orally with maximum dose of 5000 mg/kg body weight.

Groups and treatment (Each group containing 6 animals)

Group 1: Normal control (Vehicle treated, 0.1ml/100gm;*p.o*)
Group 2: Scopolamine (1 mg/kg;*p.o.)*
Group 3: Scopolamine (1mg/kg;*p.o*) + Piracetam (250mg/kg;*p.o.)*
Group 4: Scopolamine (1mg/kg;*p.o*) + EDCL (100mg/kg;*p.o*)
Group 5: Scopolamine (1mg/kg;*p.o*)+EDCL (250mg/kg;*p.o*)
Group 6: Scopolamine (1mg/kg;*p.o*)+EDCL (500mg/kg;*p.o*)
Group 7: Normal control (0.1ml/100gm;*p.o*)+EDCL (250mg/kg;*p.o*)

Preparation of Drug Solution

Preparation of Scopolamine solution:

Scopolamine was dissolved in distilled water according to the required dose and concentration. The volume of drug solution to be administered *p.o* and was calculated based upon the body weight of animals.

Preparation of Test solution:

Accurately weighed quantity of EDCL was dissolved in the distilled water to prepare the solution of the extract with appropriate concentration. The volume of drug solution to be administered *p.o* and was calculated based upon the body weight of animals.

Preparation of Standard drug solution:

Standard drug Piracetam was dissolved in distilled water according to the required dose and concentration. The volume of drug solution to be administered *p.o* and was calculated based upon the body weight of animals.

METHODS

Elevated plus Maze (EPM)

The apparatus consist of 2open arms (16 cm × 5 cm) and 2 enclosed arms (16 cm×5cm×12 cm). The arms extended from a central platform (5 cm × 5 cm) and the maze was elevated to a height of 25 cm from the floor. On the first day, each mice was placed at the end of an open arm, facing away from the central platform. Transfer latency (TL) was the time taken by mice with all its four legs to move into either of the enclosed arms. TL was recorded on the first day. If the animal did not enter into one of the enclosed arms within 90s, it was gently pushed into one of the two enclosed arms and the TL was assigned as 90 s. Retention of this learned-task was re-examined 24 h after the first day trial i.e. 2nd day. [12-14] all the treatment was given p.o.

Calculations of Inflexion Ratio(IR)

The TL was expressed as retention after 24 h by calculating the using the formula,

$$\mathbf{IR = (L1 – L0)/L0}$$

Where, L0 = transfer latency after day 7. L1 = initial transfer latency.

Morris and water maze (MWM)

The animals for the experiments were preselected by conducting at least one daily training trail. At the beginning of trail animals are place in the Morris water maze and allowed to swim freely and to escape on hidden platform. The trial was considered to be successful when mice set on the hidden platform within 3 minutes. Time spent more than 3 minutes to find hidden platform recorded as error. The percentage of successful mice was calculation as the index of Morris water maze task performance. [15] mice were dosed once in a day with the respective drugs for 10 days along with daily training trail. All the treatment was given *p.o.*30 minutes before the scopolamine treatment for 11 days and scopolamine was given *p.o.* for 30 minutes before the treatment.

Passive shock avoidance test (PAT)

Passive avoidance task based on negative reinforcement was used to examine the long term Memory. The apparatus consisted of a wooden box (27 X 27 X 27 cm) having 3 walls of wood and 1wall of Plexiglas, featuring a grid floor (3 mm stainless steel rods set 8 mm apart), with a wooden platform (10 X 7 X 1.7 cm) in the center of the grid floor. Electric shock (1Hz, 500 msec, 40V DC) was delivered for 15 sec. to the grid floor. The individual mouse was placed on the elevated platform made up of wood i.e. the shock free zone (SFZ), and the step down latency (SDL) was noted. [16-17] SDL was the time taken by the mice to step down from wooden platform. The 2^{nd} –session was carried out 9^{th} day after the 1^{st} day test. When the mice were removed from shock free zone if they did not step down for a period of 60sec. retention was tested after 24 h in similar manner, except that the electric shocks were not applied to the grid floor. Each mice was again placed on the platform, and the SDL was recorded with an upper cut- off time of 300 sec.

Calculations of Inflexion Ratio (IR)

Inflexion ratio = (L1 - L0)/L0,

Where, **L0** is the initial SDL in sec. **L1** is the SDL on the Day-9 in sec.

Object Recognition Test (ORT)

A plastic box (35cm×35cm×35 cm) and 2 identical (A and B) and 1 novel (C) object was used to study. The general procedure consisted of 3 different phases: a habituation phase, an acquisition phase, and a retention phase. Habituation phase: On the 1^{st}day mice were individually subjected to a single familiarization session of 10 min, during which they were introduced in the empty arena, in order to become familiar with the apparatus. Acquisition phase: On the 2^{nd}day animals were subjected to a single 10-min session, during which floor-fixed 2 objects (A and B) were placed in a symmetric position in the central line of the arena, 10 cm from each and 8 cm from the nearest wall. The two objects made of the same material with the similar color and smell, and identical in size. Mice were allowed to explore the objects in the open field. The exploration time on each object was shown (as seconds) to indicate the exploring activity of mice. Retention phase: On the 3^{rd}day mice were allowed to explore the open field in the presence of 2 objects: the familiar object (A) and a novel object(C) in different shapes but in similar color and size and the exploration time was calculated.

Recognition index (for retention session), calculated for each mouse, was expressed as the ratio (TC×100)/ (TA + TC), where TA and TC are the time spent during retention phase on object A and C, respectively. The time spent exploring any object (nose pointing toward the object at a distance ≤1 cm, but not mounting on the object or playing with the object) was recorded. [19, 20]

Estimation of Acetylcholinesterase(AchE) activity:

On day18^{th} day, all mice were quickly decapitated by mild Chloroform and brain was isolated from the skull immediately. The whole brain AChE inhibitory activity of extract was measured as described Ellman (1961). [21] Briefly, the whole brain is homogenized in ice cold 0.1 M phosphate buffer (pH 8.0) using Remi cooling homogenizer. The homogenates were centrifuged at 10,000 rpm for 20 min at 40^{o}C, and supernatant was used as a source of enzyme in AChE assay. The total acetyl cholinesterase activity in the aliquot of the

homogenate was estimated. The aliquot (0.3ml) was mixed with phosphate buffer (2.6ml) (pH 8.0). To this, the substrate acetyl Thiocholine iodide and dithiobis' nitro benzoic acid (DTNB) reagent were added. Acetylthiocholine iodide was hydrolyzed to Thiocholine and acetate by AChE. Thiocholine react with DTNB reagent to produce a yellow color. The rate of formation of Thiocholine from acetylcholine iodide in the presence of tissue cholinesterase was measured using a spectrophotometer. Change in absorbance per minute of the sample was read at 412 nm. The enzyme activity is expressed as the 'n' moles of substrate hydrolyzed/min/mg of protein. The protein contents in the brain sample homogenates were determined using Lowry method. [22]

$$\mathbf{R = \frac{\Delta\ O.D. \times Volume\ of\ Assay\ (3\ ml)}{E\ (13{,}600) \times mg\ of\ Protein}}$$

Where, **R**=the rate of enzyme activity in 'n' mole of acetylcholine iodide hydrolyzed /minute / mg of protein.
Δ O.D= change in absorbance/minute and
E is the extinction co-efficient, which is 13,600 M/cm.

Statistical analysis

All the observations are given as mean ± SEM, n= 6; One way ANOVA followed by Dunnett's Test. * P < 0.05; **P< 0.01; vs. respective control was considered significant.

RESULTS AND DISCUSSION

The preliminary phytochemical studies of Ethanolic extract of *Daucuscarota*Linn. Leaves(EDCL) extract shows that the presence Reducing sugar, Alkaloids, Glycosides, Flavonoids, Protein, Phenols, Amino acids, Tannins.

Animals treated with EDCL were free of any toxicity as per acceptable range given by the OECD guidelines no. 423 and no mortality was found up to 5000 mg/kg.

Elevated Plus Maze (EPM)

The Transfer Latency (TL) on EPM was expressed as IR. On the elevated plus maze the retention of learned task was studied after 24 h and at the end of day 7 as TL. Scopolamine treated mice shows increase in transfer latency and decrease in I.R. Piracetam shows significantly increased in I.R. and decrease TL. EDCL with the various doses (100,250,500 mg/kg) significantly decrease(*P<0.01*) the TL and antagonised effect of scopolamine. EDCL 250mg/kg with normal mice shows significantly decrease(*P<0.01*) inTL and increased in I.R. Observations are given in Table 1.

Table 1: Effect of EDCL on Transfer Latency (TL) on EPM in mice

Groups	Treatment	Transfer Latency (Sec)			Inflexion Ratio(I.R.)
		Day 1	Day 2	Day 7	
1.	Control (Vehicle treated)	79.50±0.57	62.16±0.63	50.12±0.61	0.55 %±0.021
2.	Scopolamine(1 mg/kg)	91.00±0.53	89.83±0.55	82.00±0.51	0.109 %± 0.003
3.	Scopol.+ Piracetam (250mg/kg)	52.00± 0.44	27.00±0.36**	19.00± 0.43**	0.68 %±0.081**
4.	Scopol.+ EDCL (100mg/kg)	63.00± 0.45	56.66± 0.51**	37.61± 0.25**	0.66 %±0.023**
5.	Scopol.+ EDCL (250mg/kg)	61.00± 0.49	45.83± 0.49**	29.00± 0.55**	1.136 %±0.034**
6.	Scopol. + EDCL (500mg/kg)	57.00± 0.67	38.00± 0.36**	25.33± 0.36**	1.26 %±0.04**
7.	Normal + EDCL (250mg/kg)	49.50± 0.73	28.50± 0.34**	17.00± 0.22**	1.19 %±0.077**

Data presented as mean ± SEM, n= 6; One way ANOVA followed by Dunnett's Test.
*P < 0.05; **P< 0.01; vs. respective Control.

Morris-Water Maze Model (MWM)

In Morris –Water Maze used for Memory Retention (MR) has found that the scopolamine treated mice shows decrease in task performance and memory retention. Piracetam group shows Memory Retention

and increase in morris-water maze task performance. EDCL with the various doses (100,250,500 mg/kg) significantly(*P<0.01*) showMemory Retentionand increase in morris-water maze task performance and antagonised effect of scopolamine. Also Normal mice treated with EDCL 250mg/kg shows significantly(*P<0.01*) increase in learning memory performance Escape Latency was decreased in EDCL treated groups due to cognitive functioning and increased in scopolamine due to cognitive impairment. The observations are given in Table 2.

Table 2: Effect of EDCL on Learning Performance of mice on MWM

Groups	Treatment	Morris- Water Maze Performance (Sec)	
		DAY 1	DAY 11
1.	Control (Vehicle treated)	52.50±0.33	42.25±0.61
2.	Scopolamine(1 mg/kg)	77.22±0.61	74.33±0.65*
3.	Scopol.+ Piracetam (250 mg/kg)	41.51±0.63	30.66±0.36**
4.	Scopol.+ EDCL (100 mg/kg)	47.15±0.42	39.16±0.42**
5.	Scopol.+ EDCL (250 mg/kg)	45.66±0.55	34.33±0.35**
6.	Scopol. + EDCL (500 mg/kg)	39.00±0.51	30.23±0.25**
7.	Normal + EDCL (250 mg/kg)	36.33±0.40	26.83±0.47**

Data presented as mean ± SEM, n= 6; One way ANOVA followed by Dunnett's Test.
*P < 0.05; **P< 0.01; vs. respective Control.

Object Recognition Test (ORT)

The R.I. of Control group was (51.33± 0.53). Scopolamine induced amnestic mice as decreases the novel object exploration time and R.I. (31.89± 0.30). Piracetam has significantly(*P<0.01*) increased the R.I. (78.16± 0.67) in presence of scopolamine. EDCL has significantly(*P<0.01*) increase the R.I. in dose dependent and EDCL Normal mice treated with EDCL 250mg/kg shows significantly(*P<0.01*) increase in novel object exploration time and shows more R.I. (78.41± 0.47).The observations are shown in Table 3.

Table 3: Effect of EDCL on Recognition Index of mice on ORT

Groups	Treatment	Recognition Index (%)
1.	Control (Vehicle treated)	51.33 ± 0.53
2.	Scopolamine (1 mg/kg)	31.89 ± 0.30*
3.	Scopol.+ Piracetam (250 mg/kg)	78.16 ± 0.67**
4.	Scopol.+ EDCL (100 mg/kg)	56.47 ± 0.53**
5.	Scopol.+ EDCL (250 mg/kg)	62.25 ± 0.34**
6.	Scopol. + EDCL (500 mg/kg)	66.50 ± 1.01**
7.	Normal + EDCL (250 mg/kg)	78.41 ± 0.47**

Data presented as mean ± SEM, n= 6; One way ANOVA followed by Dunnett's Test.
* P < 0.05; **P< 0.01; vs. respective Control.

Passive Avoidance Test (PAT)

The Step down Latency was assessed as Inflexion Ration (R.I.). Scopolamine induced amnestic mice shows decrease in SDL. Piracetam treated group shows significantly increase in step down latency in mice as compared to control and scopolamine group and EDCL with the various doses (100,250,500 mg/kg) shows significantly(*P<0.01*) increase the SDL as compared to control and scopolamine group on day 9. Normal mice treated with EDCL 250mg/kg shows significantly increase the SDL. All the Observations are given in Table.4.

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Table 4: Effect of EDCL on Step down Latency of mice on PAT

Groups	Treatment	Step Down Latency (Sec)		Inflexion Ratio (%.)
		Day 1	Day 9	
1.	Control (Vehicle treated)	36.66± 1.38	51.63± 1.10	0.40± 0.06
2.	Scopolamine (1 mg/kg)	16.33± 0.51	14.66± 0.42	-0.07± 0.04
3.	Scopol.+ Piracetam (250 mg/kg)	81.33± 1.68	119.65± 3.22**	0.47± 0.05**
4.	Scopol.+ EDCL (100 mg/kg)	51.50± 0.76	69.38± 1.92**	0.34± 0.032**
5.	Scopol.+ EDCL (250 mg/kg)	56.50± 0.68	81.58± 1.07**	0.44± 0.053**
6.	Scopol. + EDCL (500 mg/kg)	68.16± 0.32	95.50± 1.11**	0.38± 0.811**
7.	Normal + EDCL (250 mg/kg)	79.50± 0.84	112.15± 0.68**	0.42± 0.13**

Data presented as mean ± SEM, n= 6; One way ANOVA followed by Dunnett's Test.
* P < 0.05; **P< 0.01; vs. respective Control.

Estimation of Acetylcholinesterase (AchE)

The whole mice brain AchElevelin normal mice group shows concentration of AChE (146.14 ± 0.24). Scopolamine shows significant (*P<0.05*) rise in AChE activity (179.60 ± 0.91) as compared to control, piracetam and EDCL treated mice groups. Piracetam treated group shows significantly (*P<0.01*) decrese in AchE level (94.75 ± 0.31) compared to normal and amnestic mice group. EDCL with the various doses (100,250,500 mg/kg) shows significantly (*P<0.01*) lowered AchE level. EDCL 250mg/kg treated with normal mice shows significantly(*P<0.15*) decrease inAchE level (76.33 ± 0.34) as compared to normal and scopolamine treated mice.Results are shown in Figure1.

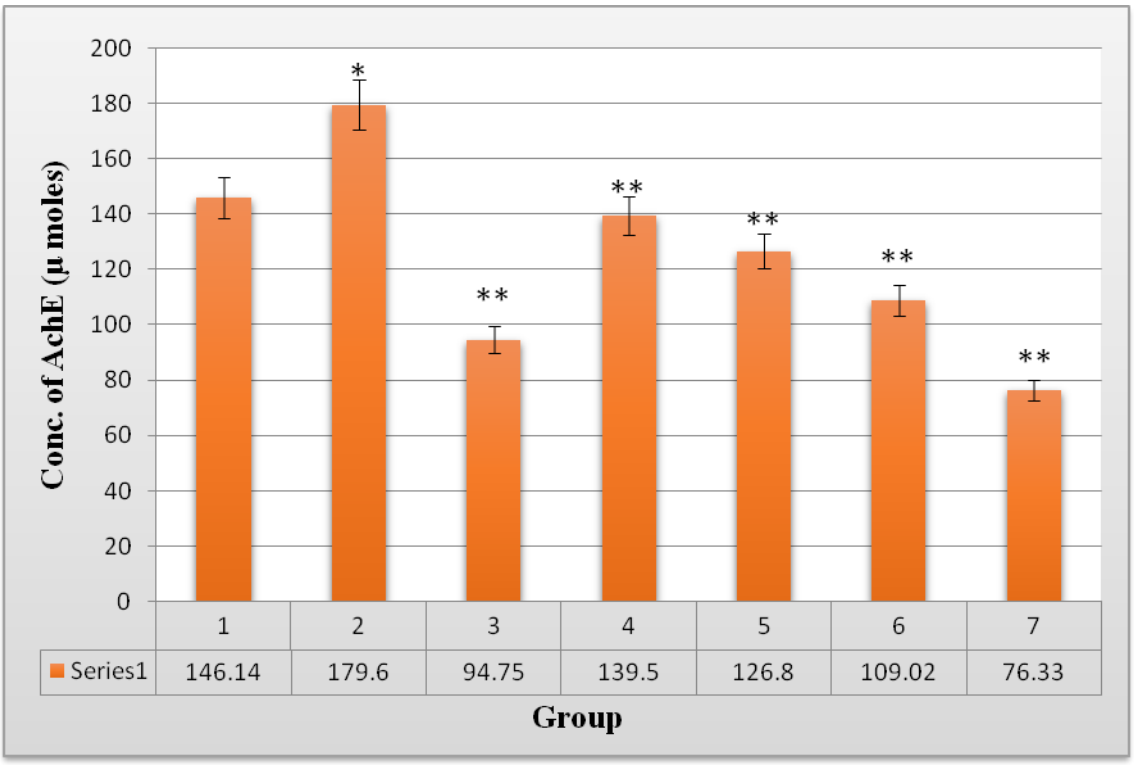

Figure 1: Effect of EDCL on Acetylcholine esterase (AchE) in mice brain.

All the observations are given as mean ± SEM, n= 6; the data was analyzed by student's *t* test. * P < 0.05; **P< 0.01; VS. Respective Control was considered significant.

DISCUSSION

The central cholinergic pathways play a prominent role in learning and memory processes. Loss of cholinergic cells particularly in the basal forebrain is accompanied by loss of the neurotransmitter Acetylcholine. Decrease in Ach in the brain of patients with AD appears to be a critical element in producing dementia. [4, 5] recently, the treatments for the dementia are acetylcholinesterase inhibitors which increase the availability of Ach at cholinergic synapses. AChE inhibitors from general chemical classes such as physostigmine, tacrine and galantamine like drugs have been tested for the symptomatic treatment of AD or dementia. [18, 19] Nootropics are the agents that enhance learning and memory. They are drugs, supplements, nutraceuticals, and functional foods that are provided improve mental functions such as cognition, memory, intelligence, motivation, attention, and concentration. [16, 17] Medicinal plants have been used to treat many psychotropic and behavioral conditions, such as dementia, Alzheimer's disease, poor memory, anxiety, seizures, insomnia, depression and drug intoxication. [20] From literature survey the *Daucuscarota*linn. Leaves show the presence of carotenoids (β-carotene) and *Daucuscarota*linn. Root also contain carotenoids (β-carotene) used regularly in Indian kitchen. In the present study we evaluated the effect of Ethanolic*Daucuscarota*linn. Leaves on learning and memory performance in scopolamine treated with mice.

Scopolamine is muscarinic receptor antagonists with amnesic properties that have been used for decades in experimental animals to induce impairment in their performance of a variety of tasks requiring intact working and reference memory. Since the first reports of a central cholinergic deficit associated with Alzheimer's disease, the connection had been made between the cognitive and memory deficits associated with this disease and the reversible amnesic effects induced by centrally acting muscarinic cholinergic antagonists. Indeed, blockade of central muscarinic receptors could induce a pattern of cognitive impairment even in young subjects reminiscent of that observed in the aged, or in individuals with Alzheimer's disease. For many years, the amnesic action produced in animals by the administration of centrally acting muscarinic cholinergic antagonists, particularly scopolamine, has been a widely used model for the characterization of potential cognition-enhancing drugs. [1-4]

The Elevated plus Maze served as the exteroceptive behavioral model to evaluate learning and memory in mice. [23] Mice show natural aversion to open and high spaces and therefore, spend more time in enclosed arms in EPM. [26] The observation and results shows that EDCL has decreased the transfer latency and increase Inflexion ratio on 2^{nd} and 7^{th} day in mice on the EPM model indicating enhancement of memory in amnestic mice. EDCL increase the cortical muscarinic acetylcholine receptor capacity. Thus, EDCL meets a major role for cognition enhancing and memory improving effects activity observed in cognitive deficit mice.

The Morris water maze has been used extensively in the study of specific test of spatial memory in mice. This is originally developed and advanced by Rich and Morris from 1981-1984. [24] Escape Latency was increased in scopolamine treated mice due to cognitive impairment. EDCL shows significantly Memory Retention and increase task performance with decrease Escape Latency on 11^{th} day in morris-water maze in scopolamine treated mice. This clearly indicates EDCL treated mice significantly increased the learning memory performance.

Passive-avoidance test based on negative reinforcement was recorded to examine long-term memory. ^{3}EDCL with the various doses shows significantly increase the step down latency and I.R. as compared to control group on 9^{th} day and antagonised effect of scopolamine due to memory enhancing effect of EDCL and decrease step down latency in scopolamine due to cognitive impairment in mice. EDCL shows improvement in passive avoidance acquisition and memory retrieval. The EDCL increased occupancy in the shock free zone (SFZ) of the paradigm and also exhibited diminished to the preferred shock zone. The increasing of step down latency by EDCL indicated in improvement in memory in the absence of cognitive deficit.

The Object Recognition Test suggests that spontaneous tendency of mice to explore the novel object (condition) in the test trial based on memory of previous experience from the acquisition trial. [19, 20] EDCL treated mice after experiencing an acquisition trial and spent more time with exploring the novel object as compared toscopolamine treated group and increase recognition index and also antagonised effect of Scopolamine in amnestic mice due to memory enhancing effect of EDCL.

Acetylcholine is considered as the most important neurotransmitter involved in the regulation of cognitive functions. There are extensive evidences linking decreased cholinesterase activity and improvement in memory.Cognitive dysfunction has been shown to be associated with impaired cholinergic functions and scopolamine is shows reduction in hippocampal Ach concentration and causes cognition impairment in mice. Selective loss of cholinergic neurons and decrease in cholineAcetyltransferase activity was reported to be characteristic feature of senile dementia in AD. [25] EDCL treated with the various doses is found to inhibit the rise in AchEactivity with the facilitation of central cholinergic activity and also improved memoryin mice brain with protected against scopolamine induced amnesia in mice. Normal mice treated with EDCL 250mg/kg groupshows a potential anti-cholinesterase activity. It also possesses nootropic activity with facilitator effect on retention of acquired learning.

Thus from results and observations EDCL has potential anti-cholinesterase agent and it enhanced memory and protected against scopolamine induced amnesia in mice shows nootropic activity in mice.

CONCLUSION

In the present study we have evaluated the Nootropic activity of EDCL with various doses in mice using Elevated plus Maze, Morris-Water Maze, Object recognition test, Passive avoidance test, Estimation of Acetyl cholinesterase (AchE) level was also done in mice brain. It was concluded that EDCL Possesses Nootropic activity which shows relation between memory improving effect and involvement cholinesterase activity and further study will also needed.

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