

Neurogenic Alterations in the pilocarpine induced epileptic activity of Wistar rats treated with Saraswata Churna

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Abstract

Approximately 80% of patients with TLE have temporal lobe epilepsy (TLE), one of the most drug-resistant seizure diseases. One of the most common signs of status epilepticus is memory impairment. Cognitive deficits are more common in patients with temporal lobe epilepsy (TLE). The findings imply that this decline in cognitive performance may be related to the development of localized brain lesions, particularly in the cortical and hippocampus areas. We generated chronic epileptic rats treated with pilocarpine in order to replicate the cognitive impairments linked to epilepsy in an animal model. We provide a protocol for two behavioral tests that are administered to epileptic rats: the Novel Object Recognition Test (NORT) to assess memory for objects, and the Morris Water Maze Test (MWMT) to assess memory for places. The purpose of the current study was to determine if the ayurvedic herb Saraswata Churna (SC) serves as a neuroprotective agent, reducing hippocampus damage and improving learning and memory performance. Using a rat model of pilocarpine-induced epilepsy, the current study examined the therapeutic effects of SC on memory and learning. Four groups were used: a normal control (NC), a pilocarpine (PI) group, a phenytoin (PHE) treated group, and a Saraswata Churna (SC) treated group. In order to investigate the potential therapeutic effects of SC for improving learning and memory, a rat model of TLE was established using lithium-pilocarpine. Our research revealed that focal brain lesions, specifically in the hippocampus and cortical regions in the SE rat model, caused cognitive abnormalities in epilepsy-induced rats. These abnormalities significantly hampered the rats' ability to learn and remember things. Preventive treatment with SC reduced learning and memory deficits by preventing neuronal cell loss and repairing hippocampal cell damage.

Keywords: Epilepsy, Pilocarpine, Saraswata Churna, Cognition, Morris Water Maze Test, Novel Object Recognition Test, Spatial Learning and Memory

Introduction

TLE is a chronic brain illness marked by a proclivity for epileptic seizures, including status epilepticus (SE) episodes[1]. Among underdeveloped nations, the prevalence of epilepsy is around 10–15 per 10,000 individuals, while it is often much greater in infants and the elderly[2]. The predominant therapeutic technique for epilepsy is pharmacological intervention; however, roughly two-third of epileptic patients who are resistant to the antiseizure drugs [3]. Furthermore, currently existing antiepileptic medicines (AEDs) are only effective at suppressing seizure symptoms and not at avoiding the underlying pathology of epileptogenesis[4]. As a result, effective therapeutics targeting the molecular and cellular mechanisms underlying the epileptogenic process are desperately needed[5]. Numerous diagnostic and therapeutic, pathophysiological, and pharmacological studies have conclusively demonstrated that chronic inflammation processes in the cerebral cortex and hippocampus play a vital role in hippocampal neurogenesis[6]. Autoimmune therapies are typically more effective than AEDs in the treatment of TLE because the defense system is activated in the neurons of TLE patients[7]. In a rat TLE experimental model, similar observations were also verified[8].

Because repeated seizures can create morphological and chemical changes in brain activity, aberrant seizure activity can contribute to cognitive impairment, which is one of the most common epilepsy-related disorders [9-11]. In contrast to chronic seizure occurrences, which are transitory and momentary, cognitive deficits can persist across seizure individuals' lifespan, affecting their quality of life [12]. As a result, it's critical to

comprehend the pathophysiologic mechanisms behind epilepsy-related cognitive deterioration. A variety of experimental animal models of epilepsy have been used to highlight the memory and cognitive impairments associated with chronic epilepsy[13]. Memory loss in TLE patients has been assessed using the Morris water maze, contextual fear conditioning, hole-board, new object location (NL), and novel object recognition (NORT) tests, among others[14]. Since the hippocampus is among the most prevalent sites where TLE develops, behavioural tests that evaluate hippocampal-dependent memory function are widely used[15]. Behavioral paradigms for assessing dentate neonatal neuronal function can also disclose critical data regarding the biological mechanisms behind epilepsy-related memory problems, because seizures can produce aberrant cortical formation and contribute to epilepsy-related neurodegeneration which will cause the cognitive impairment [16].

We explain how epileptic rats can execute a variety of memory tests, including the NORT and the MWM Test, in this study. The tests are simple and straightforward, and they do not necessitate the use of a complex system.

Materials and Methods:

Animals

Four month old adult male Wistar rats (200 - 210 g; n -total = 24) obtained from Central Animal House Facilities in Manipal Academy of Higher Education, Karnataka, India with Ethical approval. All the animals were housed in groups of 3 rats per cage under controlled conditions of luminosity (12-hour dark-light cycle), humidity (55%) and temperature (20 ± 2 °C), with food and water *ad libitum*.

Lithium-Pilocarpine model of epilepsy

Animals were intraperitoneally injected with pilocarpine (270 mg/Kg, i.p.) 18-24 hours after lithium injection (127 mg/Kg, i.p.). The behavioral scores of seizures were recorded according to the Racine's scale (Racine, 1972). One hour after pilocarpine injection, SE was terminated by injection of diazepam (10 mg/Kg, i.p.). The normal control group animals were aged and left untreated.

Experimental groups

All the 24 rats were divided into four groups ($n= 6$ /group) as Normal Control group (NC), Pilocarpine (PI), Phenytoin (PHE) and Saraswata Churna (SC) group. Epilepsy model was created by a single intraperitoneal injection (270mg/kgbw) of pilocarpine. At the end of 24 hours and 48 hours post first seizure occurrence, Phenytoin 30mg/kgbw (i.p.) and SC (308 mg/kgbw oral) were given to the respective groups. After 8 days of experimental period, learning and memory of rats were evaluated by NORT and MWM test.

NORT – for Evaluating the Cognition:

This assessment is based on rodents' natural tendency to visit a novel object more frequently than a known object. The preference for examining novel objects is a behavioural measure of remembering and discriminating between known and novel objects, demonstrating the hippocampus's learning and memory function. The technique is usually divided into three stages: habituation, acquisition, and testing. The rat is permitted to habituate in the open field arena for 30 minutes during habituation. Two identical objects are placed in the open field during acquisition (Trial 1), and the rat is permitted to examine the objects for 5 minutes before being returned to their home cage. After twenty-four hours, the rat will be returned to the open field arena, which contains both familiar and unknown things (Trial 2- Test Phase). The number of trips to the unique object and the amount of time spent near it will be scored (Ennaceur & Delacour 1988). The entire experiment will be recorded on videotape for later analysis.

Outcome Measures:

Score the first 5 minutes in both Trial 1 and Trial 2. Continue scoring the past 5 minutes until total exploration time reaches 20 seconds, if the rat does not meet the minimum exploration duration of 20 seconds for both the objects.

MWM Test for Assessing Spatial Learning and Memory:

It's a hippocampal-dependent exercise that's also the most extensively utilised paradigm for evaluating memory and spatial learning[17]. MWM apparatus had a large circular pool (1.5 mts diameter) divided into four imaginary quadrants and filled with water with a temperature of 19-23°C with a depth of 40 cms. An island (hidden platform) was immersed 1 cm under the surface of water in one of the four zones/quadrants which is considered the target quadrant. This hidden platform is 4" × 4" size and camouflaged with non-toxic white tempura paint to make the water opaque[17-19]. It is referred to as a 'maze' in the sense that the animals need to discover the hidden platform, which is a small goal located in a fixed position and immersed below the water

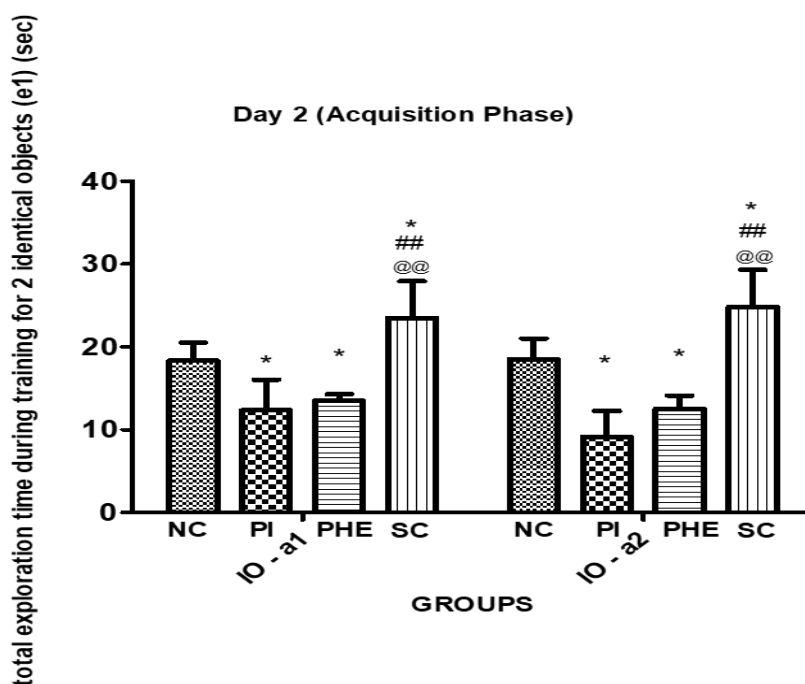
surface. In case the animal did not reach the hidden platform within 60 seconds, it required guidance to reach the platform and was permitted to stay on the platform for 20 seconds[20]. The entire apparatus was divided into five zones. The system does not track the head movements. It also does not detect immobility whilst tracking the animal. It records the rat's position at most 10 times per second. While tracking, the system displayed the time remaining in the test and marked the position of the rat's centre of gravity. A video camera (Logitech B525 HD Webcam) connected to a computer system (HANNES-G) was placed above the centre of the pool which uses the entire captured image (640×480 pixels). This computer system had a special tracking software (ANY-maze version 4.82) to track the movement of the rats. The rats were habituated for 5- 10 minutes in the testing room, as immediate trial will lead to over excitement of the animals[17, 18]. Following their exposure, the animals were trained for five days in a row, with four trials each day for each animal. The rats were positioned in various starting positions towards the pool's side walls during each session. The time it took to reach the concealed platform at the conclusion of each trial was recorded[20]. On the sixth day, post the last training session the animals were subjected to one session of memory retention test where the hidden platform was removed. Memory retention test was performed for 45 seconds. The total time taken by each animal to reach the target quadrant during the four consecutive training days and memory retention test was calculated in seconds. Briefly, the rats were subjected to 5-day learning and memory training sessions followed by a memory retention test on the 6th day. In order to reduce the excitement of strange environment, all the animals were subjected for a trial on a day before the actual training sessions. The computer generated data was analysed and compared across all the groups, with standard comparison to control group.

Results

Novel object recognition test (NORT):

Acquisition Phase - Total Time Explored During Training for 2 Identical Objects:

The time taken to explore the IO-a1 on day 2 (Acquisition Phase) of the three days experimental period of the NORT by the animals are: Normal Control (NC) group (36.7%), Pilocarpine Group (PI) group (24.7%), Phenytoin (PH) treated group (27.0%), and the drug Saraswata Churna (SC) treated group (47.0%) and the time taken to explore the IO-a2 by the Normal Control (NC) group (37.0%), Pilocarpine Group (PI) group (18.3%), Phenytoin (PH) treated group (25.0%), and the drug Saraswata Churna (SC) treated group (49.7%). The saraswata churna treated group shows very significant difference ($p < 0.01$) and the pilocarpine and phenytoin group shows mild significance ($p < 0.05$) in exploring and identifying the two Identical Objects (IO-a1 and IO-a2), Graph 01. This result shows that SC treated group spent more time in exploring the Identical Object IO-a1 and IO-a2 in comparison to the PI and PH group which shows the cognitive effect of SC is significant in learning and memory.



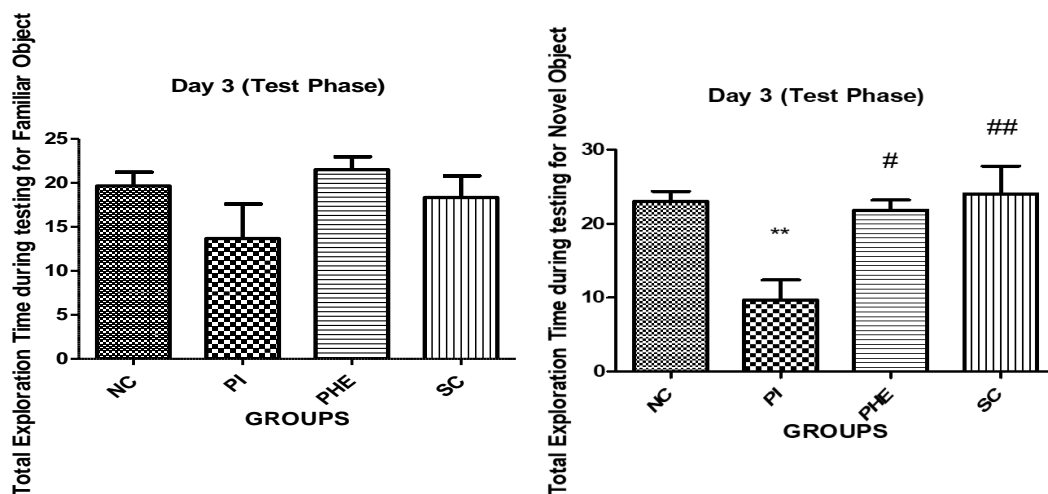
Graph: 01. The above graph shows the comparison in time taken to explore the two identical objects (IO-a1 and IO-a2) on Day 2 (Acquisition Phase) by the animals in all the four groups:

Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group, IO-a1- Identical Object a1, IO-a2 – Identical Object a2, *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Test Phase - Total time taken by the rats to explore the familiar object (a) and the novel object (b) (sec) during testing phase:

The time taken to explore the familiar object on day 2 (Acquisition Phase) of the three days experimental period of the NORT by the animals are: Normal Control (NC) group (39.3%), Pilocarpine Group (PI) group (27.3%), Phenytoin (PH) treated group (43.0%), and the drug Saraswata Churna (SC) treated group (36.7%) Graph 02, and the time taken to explore the novel object by the Normal Control (NC) group (46.0%), Pilocarpine Group (PI) group (19.3%), Phenytoin (PH) treated group (43.7%), and the drug Saraswata Churna (SC) treated group (48.0%). The SC treated group spent more time in exploring the novel object and showed very significant difference ($p \leq 0.01$) in identifying the novel object in comparison to identifying the familiar object. The pilocarpine treated group spent very less time in exploring the novel object and showed very significant difference ($p \leq 0.01$) Graph 03. This result shows that SC treated group spent more time in exploring the novel Object more than the familiar object in comparison to the PI and PH group which shows the cognitive effect of SC is significant in learning and memory.



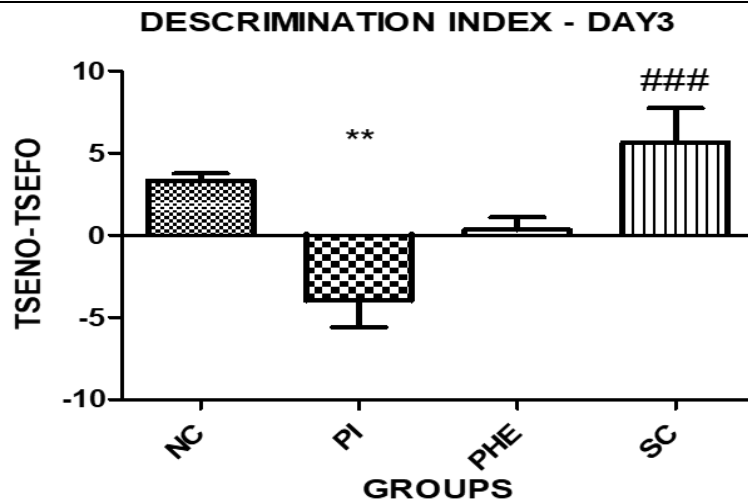
Graph: 02. The above graph shows the comparison in time taken to explore the Familiar Object on Day 3 (Test Phase) by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

Graph: 03. The above graph shows the comparison in time taken to explore the Novel Object on Day 3 (Test Phase) by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
*- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Test Phase - The absolute discrimination measure i.e., time spent by the animals to explore the novel object minus time spent to explore the familiar object (TSENO-TSEFO) on Day 3:

The SC treated group shows a highly significant difference ($p \leq 0.001$) in measuring the absolute discrimination index in comparison to the phenytoin and pilocarpine treated group and the pilocarpine treated group spent less time in exploring the novel object and showed very significant difference ($p \leq 0.01$) in measuring the absolute discrimination index showing that SC treated group performs well in retaining their memory and discriminating between the novel object and familiar object, Graph 03.

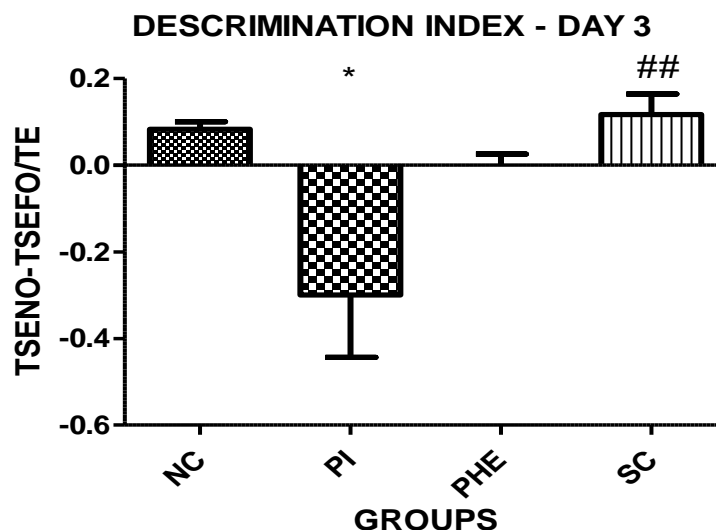


Graph: 03. The above graph shows the Discrimination index .i.e., time spent exploring the novel object minus time spent exploring the familiar object (TSENO-TSEFO) on Day 3 (Test Phase) by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Test Phase - relative discrimination index .i.e., time spent by the rats to explore the novel object minus the time spent in exploring the familiar object divided by total exploration (TSENO-TSEFO/TE) on Day 3:

The SC treated group shows good response in discriminating between the familiar object and novel object and shows a very significant difference ($p \leq 0.01$) in measuring the relative discrimination index which is between -1 and 1 in comparison to the phenytoin and pilocarpine treated group and the pilocarpine treated group spent less time in exploring the novel object and showed a significant difference ($p \leq 0.05$) in measuring the discrimination index showing that SC treated group performs well in recognising the object and retaining their memory, Graph 04.

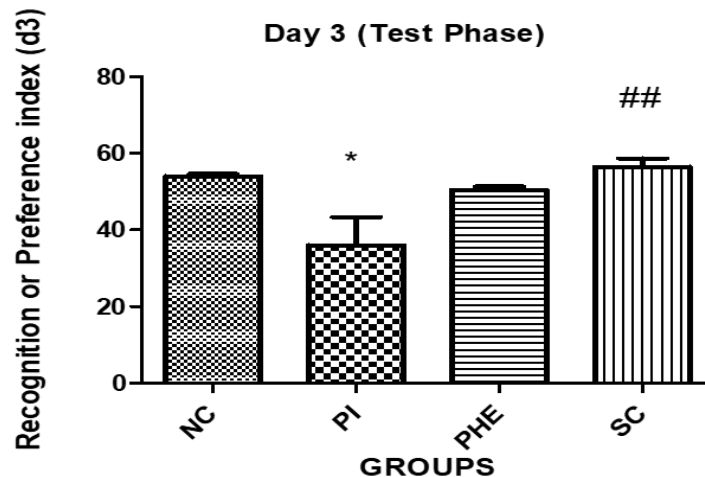


Graph: 04. The above graph shows the Discrimination index .i.e., time spent exploring the novel object minus the time spent exploring the familiar object divided by total exploration (TSENO-TSEFO/TE) on Day 3 (Test Phase) by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Test Phase - Recognition or preference index .i.e., the time spent by the rats in exploring the novel object divided by the total time on Day 3:

The SC treated group shows good response in recognising the novel object and familiar object and shows a very significant difference ($p \leq 0.01$) in measuring the preference or recognition index which is between 0 and 1 in comparison to the phenytoin and pilocarpine treated group and the pilocarpine treated group spent less time in recognising the novel object and showed a significant difference ($p \leq 0.05$) in measuring the discrimination index showing that SC treated group performs well in recognising the object and retaining their memory, Graph 05.



Graph: 05. The above graph shows the recognition or preference index (d3) .i.e., the time spent exploring the novel object divided by the total time on Day 3 (Test Phase) by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

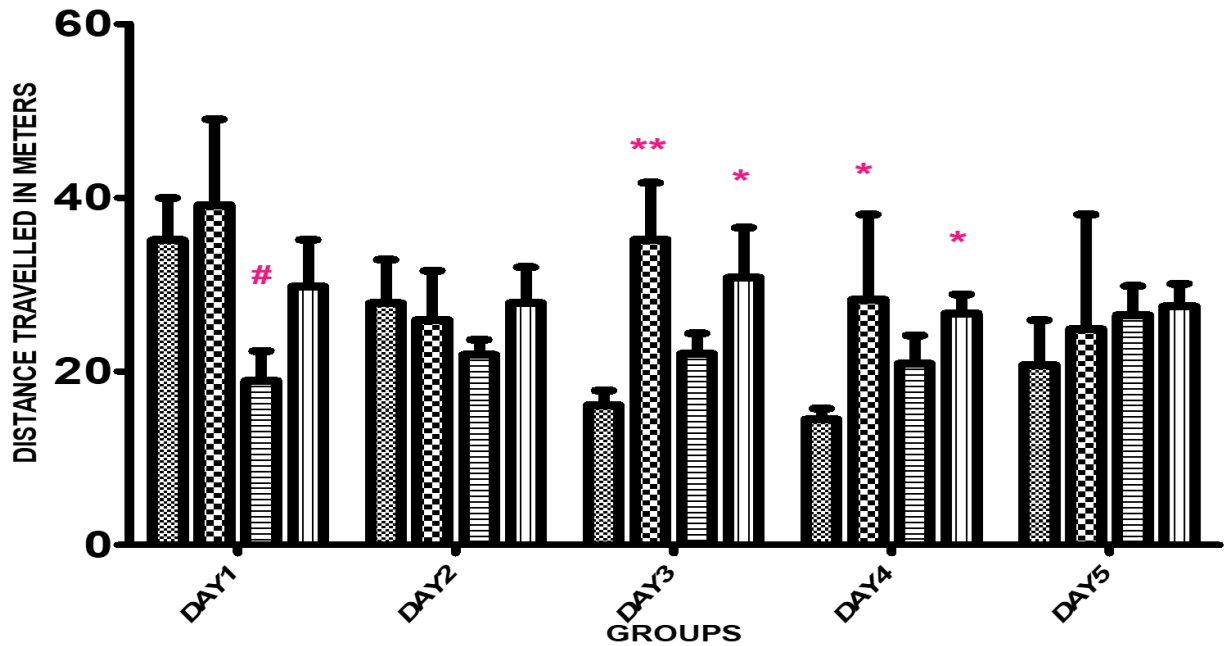
NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group

*- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Morris water maze (MWM) Test:

Overall escape latency analysis:

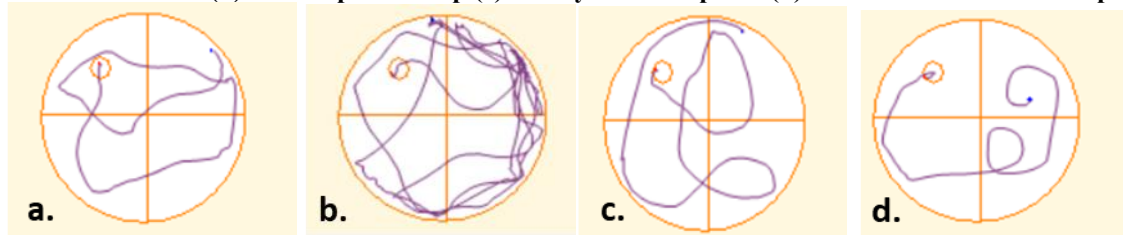
The overall escape latencies were significant between groups during day1 to 5 of training sessions in MWM day 1, $p < 0.05$ between the phenytoin and Saraswata Churna group; day 3, $p < 0.01$ between the Pilocarpine and Normal Control group as well as between Saraswata Churna and Normal Control group; day 4, $p < 0.05$ between the Pilocarpine and Normal Control group as well as between Saraswata Churna and Normal Control group, Graph.06. Post-hoc test revealed there were no significant differences on the Day 2 and Day 5. However, on the day 1, 3 and 4 of training, the Saraswata Churna treated group had significant longer latency than the control group ($p < 0.05$) Graph.07, Fig.1



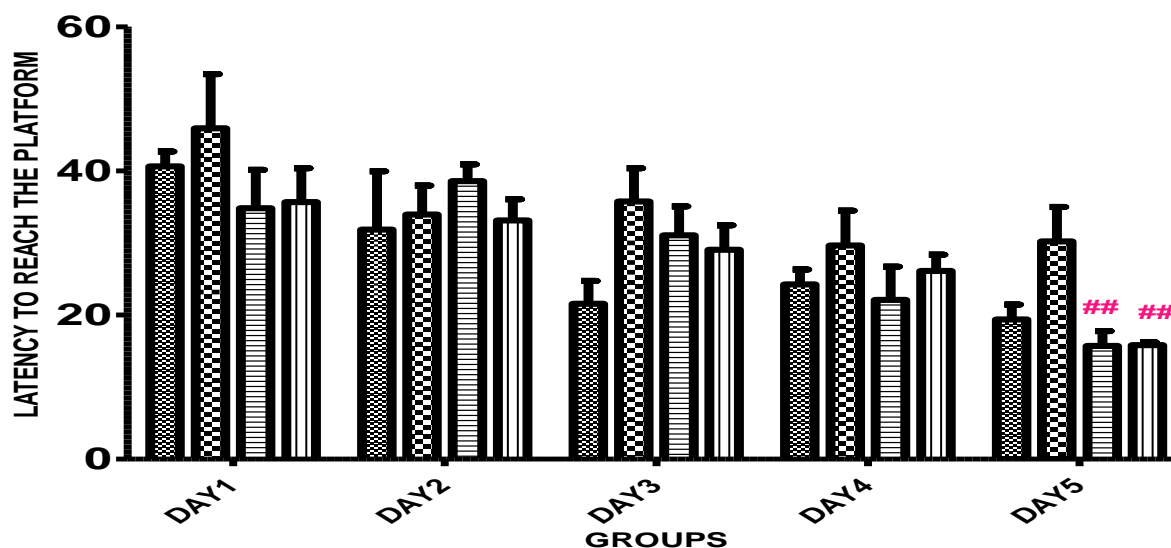
Graph: 06. The above graphs show the distance travelled (in meters) during the spatial acquisition trials on day 1-5 by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Fig.1. Track plot (using tracking software (ANY-maze version 4.82) representing the pathway taken by the animals to reach the hidden platform on all the consecutive trial days across all the four groups. (a). Normal Control (b). Pilocarpine Group (c). Phenytoin Group and (d). Saraswata Churna Group



Spatial Acquisition Trial- Day1 to Day5



Graph: 07. The above graphs show the Latency (in seconds) to reach the platform during the spatial acquisition trials on day 1-5 by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

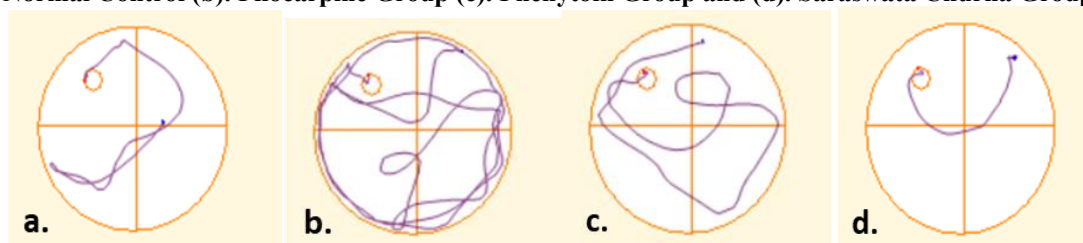
Memory Retention Test - Treatment with Saraswata Churna attenuates cognitive deficits caused by pilocarpine induced seizure:

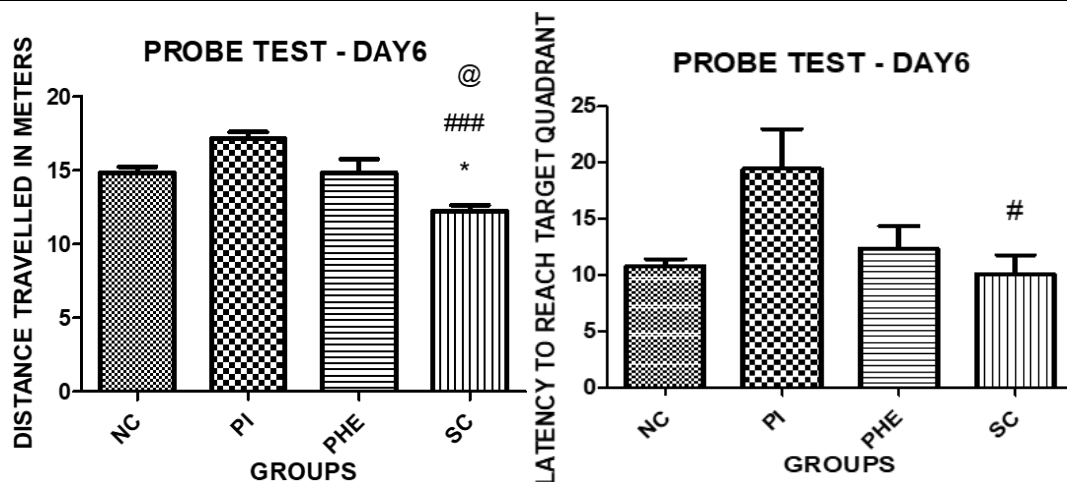
Significant main effect of Saraswata Churna were present during the sixth day of the six days training period of MWM test, in which rats of Saraswata Churna treated group showed significant decrease ($p < 0.001$ in comparison to the pilocarpine group, $p < 0.05$ when compared with the Phenytoin and Normal Control group) in the distance travelled to reach the platform Graph 08. And also showed significant decrease in the latency ($p < 0.05$ when compared with the Phenytoin and Normal Control group) to reach the escape platform, i.e. lesser duration to reach the platform, compared to the Pilocarpine and Phenytoin treated group in the memory retention test, with removal of platform, Graph 09.

Similarly the rats of Saraswata Churna treated spent more time in the target quadrant and group showed significant difference $p < 0.05$ in comparison to the pilocarpine group and Phenytoin treated group. The pilocarpine induced group showed significant decrease in the time spent in the target quadrant ($p < 0.05$) in comparison to the Phenytoin and Saraswata Churna treated group Graph 10. This results suggest that the Saraswata Churna treated group shows good memory retention Fig.2.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Fig.2. Track plot (using tracking software (ANY-maze version 4.82) representing the pathway taken by the animals to reach the hidden platform during the memory retention test across all the four groups. (a). Normal Control (b). Pilocarpine Group (c). Phenytoin Group and (d). Saraswata Churna Group

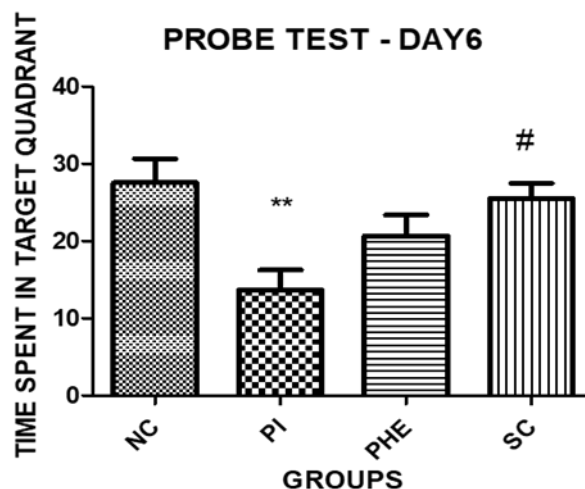




Graph: 08. The above graphs show the distance travelled (in meters) during the probe test on day 6, by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

Graph: 9. The above graphs show the Latency (in seconds) to reach the target quadrant during the probe test on day 6, by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group



Graph: 10. The above graph shows the time spent (in seconds) in the target quadrant during the probe test on day 6, by the animals in all the four groups: Normal Control (NC), Pilocarpine group (PI), Phenytoin treated group (PHE), and the drug Saraswata Churna (SC) treated groups.

NC- Normal Control Group, PI- Pilocarpine Group, PHE- Phenytoin Group, SC- Saraswata Churna Group
 *- In Comparison with NC Group, #- In Comparison with PI Group, @- In Comparison with Phenytoin Group

Discussion

The methodologies for measuring cognitive function in rats with persistent epilepsy are described in this paper. Learning and memory skills in rats are assessed using a variety of behavioural test paradigms[21]. The MWM Test, NORT, and Novel Object Location Tests are three of the most effective and standard methodologies for evaluating epileptic rats' memory and learning [22, 23]. Because epileptic rodents can have spontaneous seizures during behaviour sessions, it's best to use based on behavioral tests that are focused on the animals' natural

desire to explore novelty without adding additional positive or negative reinforcements, such as those used in aversive-motivated tasks like fear conditioning, mild starvation, or forced swimming to stay afloat, which can cause recurrent seizures[24, 25].

Saraswata Churna has a cognitive and nootropic effect on pilocarpine-induced seizures, according to the results of this study. It also appears to help with memory retention and attenuates cognitive impairments. This finding is significant in the field because it could lead to the development of a preventive therapy for people who are at risk of developing behavioural comorbidities like depressive-like behaviour, motor hyperactivity, reduced exploratory behaviour, and cognitive impairments as a result of chronic epileptic seizures.

The NORT and MWMT were used to assess cognitive impairment. Indeed, pilocarpine causes a cumulative deterioration in this test, to the point that animals are unable to distinguish between the novel and the familiar object in the chronic period, spending the same amount of time exploring each [26, 27]. Rats given Saraswata Churna during latency, on the other hand, showed no signs of impairment in this test. Because direct intra-hippocampal BDNF treatment attenuated cognitive impairment in the pilocarpine mouse, BDNF release may delay the return of memory losses [28]. Memory processing is known to be exclusive to the hippocampus and parahippocampal area [29, 30].

In pilocarpine-treated epileptic rats, cognitive deficits in spatial working memory testing were consistently revealed, with substantial hippocampal neuronal death, but object recognition memory tests yielded inconsistent findings, with varied neuronal degeneration in parahippocampal brain regions[31-33]. These findings show that, unlike spatial memory, object identification may involve intricate networking links between various regions of the brain, rather than the hippocampus playing a crucial role [12, 34-37].

Saraswata Churna exerted potential effects in ameliorating anxiety and restoring non-spatial recognition memory impairment induced by pilocarpine. This potential effect of Saraswata Churna is due to its active ingredients like Brahmi (*Bacopamonnieri*), Shankhpushpi (*Convolvulus pluricaulis*), Vach (*Acoruscalamus*), Gorakmundi (*Sphaeranthusindicus*) and Satavari (*Asparagus racemosus*)[38]. The combination has features like "brain tonic," "memory booster," and "general health tonic." Medhya Rasayana, or nootropic herbs, include Brahmi, Vacha, and Shankhpushpi. Memory enhancers, brain tonics, and cognitive enhancers are all terms used to describe nootropics. The term "cognitive enhancer" refers to a drug that improves cognition and focus. Cognitive enhancers are thought to work by altering the availability of neurochemicals in the brain, promoting neuron formation, or improving brain oxygen flow. Memory herbs improve memory and brain function by raising neurotransmitter receptors, particularly cholinergic, and enhancing blood flow to the brain through increasing oxygen and nutrient supply[39].

Brahmi comprise of the alkaloid glutamic acid, triterpenoidsaponins, nicotine, brahmine, stigmasterol, aspartic acid, herpestine, bacosides A and B, saponins A, B and C, β -sitosterolbetulinic acid, serine, D-mannitol, stigmaterol and α -alanine [40]. It has long been used to cure a variety of mental problems, as well as to aid digestion, increase learning, focus, and memory, Dementia, bronchitis, and convulsions [41-43].

The nootropic herb *Bacopa monnieri* aids in the mending of injured neurons, neuronal synthesis, synaptic activity restoration, and better brain function. Several studies have shown that the phytochemical contents of *B. monnieri* (ie, bacosides) protect the brain from oxidative damage and age-related cognitive loss in a variety of ways[44-46]. A clinical trial indicated that an oral treatment with *B.monniieri* improved memory in both adults and children[47].

Shankhpushpi (*Convolvulus pluricaulis*), a perennial herb [48], containing constituents like D-glucose, maltose, rhamnose, sucrose, volatile oils, fatty acids, fatty alcohols, hydrocarbons, and many steroids [49], is a well-known Ayurvedic drug that has been used as a memory booster, nervine tonic and gastrointestinal laxative for thousands of years. [50, 51]. This herb is used as a memory enhancer, a psychostimulant, and a tranquillizer, as well as a stress reliever[52].

Sweet flag (*A.calamus* (L.)) is a member of the *Acoraceae* family and is extensively used in Indian and Chinese traditional medicine, either alone or in combination with other herbs[53, 54]. The oil contains varying concentrations of asarone, calamenenolcalamene, pinene, eugenyl acetate, methyl isoeugenol, camphor, aldehydes, fatty acids and hydrocarbons[55-57][58]. Pneumonia, influenza, bronchitis, psychosis, seizures, anxiety, sleeplessness, emotional disturbance, dermatitis, peptic ulcers, gastroenteritis, colitis, renal and hepatic diseases have all been treated with *A.calamus*. [59]. The oil's AChE-inhibitory activity can be attributed to -

asarone. Because acetylcholine levels are linked to memory and cognitive performance, the AChE-inhibitory activity of this herb could explain its traditional use[60].

Conclusion:

In conclusion, while memory and cognitive deficits have been found in Pilocarpine-treated rats throughout the chronic phase of epileptogenesis, there have been fewer studies that have examined cognition early following Status Epilepticus. Long-term recognition memory is diminished in pilocarpine models. Our findings showed that Saraswta Churna might reduce brain inflammation following SE induction, suggesting that it could be a promising novel anti-inflammatory medication for epilepsy treatment. Additional researches using this knowledge to investigate psychologically treatments are designed in these frameworks, especially if they use the NORT and MWM Tests, are simple, quick assays that offer the rodents little distress, making them appropriate for testing cognitive performance in epileptic animals without generating periodic episodes. We hypothesize that Saraswta Churna, which has been demonstrated to have neuroprotective properties, could be used to prevent and treat neuroinflammatory disorders such as TLE. This sheds light on the link between behavioural changes and the length of an epileptic episode. All of these findings could be relevant in future studies looking into Saraswta Churna's probable neuroprotective pathways in epileptogenesis.

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