



Bioscene

Bioscene

Volume- 21 Number- 02

ISSN: 1539-2422 (P) 2055-1583 (O)

www.explorebioscene.com

The Effects of Consumption of Cashew Nut (*Anacardium Occidentale*) Diet on Scopolamine Impaired memory and Motor Coordination in CD1

*Johnbull Martins Uket¹, Christiana Godwin Gekpe¹, Helyn Andonimye Ikem¹, Peter Cecilia Agbo¹ & Nkwa Oge²

¹Department of Physiology, Faculty of Basic Medical Sciences, College of Medical Sciences, University of Calabar, Nigeria

²Department of physiology, faculty of medical sciences, Abia state university, Uturu , Nigeria

Correspondence Author : Johnbull Martins Uket

Abstract: Alzheimer's disease, the most common clinical condition with dementia is on the increase worldwide. Long term consumption of cashew nut diet has been reported to improve memory in normal animal models and this was attributed to the memory enhancing effects of tryptophan, vitamins B6 and Docosahexaenoic acid (DHA) present in cashew nut. However, whether consumption of cashew nut diet will also improve memory and motor co-ordination in mice with impaired memory and motor deficits has not been previously ascertained. Therefore, this research was to study the effects of consumption of cashew nut (*Anacardium occidentale*) diets on scopolamine-impaired memory and motor co-ordination in CD1 mice. Twenty-eight (28) experimental mice were randomly assigned into 4 groups namely; control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only groups. Feeding was done for 21 days before learning/memory and motor coordination were studied. The Morris water maze was used to estimate learning/memory and beam walking test was used for study of motor co-ordination. Results from the Morris Water maze shows that, learning and memory were significantly impaired in the group administered scopolamine only compared to all other experimental groups ($p < 0.05$). The result of the beam walking shows improved motor coordination in the groups fed cashew nut diet compared to scopolamine only group ($p < 0.05$). In conclusion, consumption of cashew nut diets improved learning and memory as well as motor coordination in mice. The memory and motor coordination enhancement effects observed may be attributed to tryptophan, DHA and vitamin B6 present in cashew nut diet.

Keywords: Alzheimer's disease, motor coordination, cashew nut, scopolamine, tryptophan, hippocampus, vitamin B6

Introduction

Neurodegenerative disorders affect the status and well-being of an individual. These abnormalities occurring largely in the brain, spinal cord or nerves can lead to varieties of symptoms ranging from learning and memory deficits (dementia), muscle

weakness, poor motor coordination, as well as cognitive decline [1]. Iskushnykhetal.,(2024) reported an observation of diminished cerebellar function in patients with neurodegenerative disorders such as Alzheimer's disease (AD), dementia, Parkinson's disease (PD), and others [2]. Alzheimer's disease is the most common type of dementia, accounting for 60 to 80% of all cases of dementia [3]. Animal studies showed that, dementia a form of Alzheimer's disease is associated with motor coordination and balance [4]. Brain centers involve in learning, memory, motor coordination and balance include: The hippocampus [5], basal ganglia [6], cerebellum [7], prefrontal cortex [8] as well as the somatosensory cortex [9].

Cashew nut (*Anacardium occidentale*) of the family anacardium is one of the tropical fruits produced globally in large scale in some parts of Brazil, India and Vietnam [10]. Cashew nut diets are nutraceuticals. The seed serves in most parts of the world as snacks or incorporated in a variety of food as source of nutrients such as protein, carbohydrate, fiber, mineral and water [11]. It has been reported that, Cashew nut have antimicrobial, antidiabetic and antioxidant effects [12], packed with essential nutrients such as vitamin B6, vitamin D, protein and other essential amino acids (tryptophan) which acts as natural antidepressants [13], as well as omega -3 fatty acids which is important for neural membrane functions in brain tissues [14]. Docosahexaenoic acid (DHA) is the highest important omega-3 fatty acid in neural membrane tissues of brain with protective effects after cellular complications [15]. De Melo *et al.*,(2017) reported that, consumption of cashew nut diet during pregnancy and lactating periods in animal models influenced the development of certain reflex responses in newborns and improve performance on memory tasks in adolescent experimental animals [16]. Notable among the array of chemical constituents present in cashew nut is serotonin precursor (tryptophan), that had been reported to influence memory and learning [17]. The relevance of serotonin in the management of locomotor activities in mammals has been a focal point in recent studies. Serotonergic pathways are included in the reticulospinal (RS) pathways activated by the locomotor signals from higher structures of the brain [18]. DHA, and Vitamin D3 had been reported to have Improved Memory in Aluminium-Induced Cognitive Impairment in Rat Models [19].

Scopolamine a natural or synthetically produced tropane alkaloid and anticholinergic drug that is used as a medication to treat motion sickness, postoperative nausea and vomiting [20], is a drug of choice in inducing memory impairment in animals including mice because, the cognitive dysfunction or memory impairment observed after this drug's usage is analogous to observations in demented patients. As a muscarinic receptor antagonist, scopolamine impairs long term potentiation which is responsible for long term memory [21]. Acetylcholine (ACh) is a parasympathomimetic neurotransmitter that can signal through ligand-gated cation channels (nicotinic receptors) and G-protein-coupled muscarinic receptors (mAChRs). ACh signalling through mAChRs located in the central nervous system (CNS) and periphery can regulate smooth muscle contraction, glandular secretions, heart rate, and various neurological phenomena such as learning and memory [22,23]. mAChRs is divided into muscarinic Acetylcholine receptors (M1-M5) five subtypes, present at various levels of the brain [23]. Scopolamine acts as a non-selective competitive inhibitor of M1-M5 mAChRs, as such, it has been classified as anticholinergic at several dose-dependent

therapeutic and adverse effects [24,25]. Recent reports and evidence suggested that M1 (and possibly M2) mAChR antagonism at interneurons acts through inhibition of downstream neurotransmitter release and subsequent pyramidal neuron activation to mediate neurological responses associated with stress and depression [26]. Similar antagonism of M4 and M5 receptors is associated with potential therapeutic benefits in neurological conditions such as schizophrenia and substance abuse disorders [23].

Considering the high cost of living globally and the adverse effects of synthetic drugs, there is a search for natural remedies that are cheaper, safer and effective pharmacologically for humanity. According to World Health Organization statistical report, 80% of the world's population presently uses traditional medicine for some aspects of primary health care including mental health [27]. Therefore, natural products may provide a new source of beneficial neuropsychotropic drugs provided they had been scientifically validated and their mechanisms properly established. Since cashew nut nutraceuticals with bioactive ingredients such as tryptophan, vitamins B6, and D3, as well as DHA that can affect learning, memory, motor coordination and balance it is, therefore, evident that, the consumption of cashew nut diet may affect memory and motor coordination. Therefore, this study is to investigate the effects of cashew nut (*Anacardium occidentale*) diet consumption on scopolamine impaired memory and motor coordination in CD1 mice.

Materials and Methods

Preparation of cashew nut diet: five (5) bottles of cashew nut were bought from Cost-low, a local mini market in Calabar, Nigeria. The cashew nuts were air dried and grounded into powdered form using an electric blender. The powdered form weighed 4,280g.

Experimental animals and design: TwentyEight (28) experimental adults CD1 mice both male and female sexes weighing 17 – 26g body weight were obtained from the animal house of Physiology Department, faculty of basic medical sciences, University of Calabar, Nigeria., were used for this study. The animals were assigned randomly into four (4) experimental groups of 7 mice in each group namely; control, cashew nut diet only, cashew nut diet plus scopolamine and scopolamine only group groups. The control group was constituted by four males and three female mice, the cashew nut diet only group was constituted by four males and three females, the cashew nut diet plus scopolamine group was constituted by four males and three females and the scopolamine only group was constituted by four males and three females. The animals were housed individually in plastic cages with wired screen top illuminated on a 12-hour light-dark cycle. The animals were allowed access to clean water and food *ad libitum*. The animals were allowed to acclimatize for 72 hours. Approval for the use of laboratory animals was obtained from the faculty of basic medical sciences, college of medicine Ethical Committee of University of Calabar, Nigeria on the use of experimental animals and it was in accordance with the internationally accepted principles for laboratory animal use and care as found in the European Community guide lines (EEC Directive of 1986; 86/609/EEC).

The mice in the control group were fed with normal rodent chow and administration of normal saline (1ml/kg bodyweight intraperitoneally). The cashew nut diet group were fed with cashew nut diet and normal saline (1ml/kg bodyweight intraperitoneally). The cashew nut diet plus scopolamine group were fed with cashew nut diet and administered scopolamine (1mg/kg bodyweight intraperitoneally) and the scopolamine only group were fed normal rodent chow and administered scopolamine (1mg/kg bodyweight intraperitoneally). All the experimental animals in the four groups were allowed access to free water and respective diet *ad libitum*. Scopolamine was administered once daily for the first week. In the subsequent weeks, Scopolamine was administered once every two days. The feeding was done for Twentyone (21) days before laboratory test that lasted for nine (9) days was done to assess learning and memory as well as motor coordination and balance.

Behavioural Protocols

Morris water maze for learning and memory: The Morris Water Maze is a behavioral task to test hippocampal-dependent learning and memory. It has been widely used in the study of neurobiology, neuropharmacology and neurocognitive disorders in rodent models [28]. Our maze model is smaller than the type developed for rats. The water maze is constructed out of a circular polypropylene pool (Canadian tire “Pelican” pool) that measures 110-cm in diameter and 20-cm in depth. The pool was filled to a depth of 14-cm (0.5-cm over the platform) with room-temperature tap water, which was made opaque with the addition of 100-mL of non-toxic white liquid tempura paint (Schola, Marieville). The water was left to sit overnight in order to attain room temperature ($22 \pm 1^\circ\text{C}$).

The pool was divided into four quadrants: Northwest, Northeast, Southwest and Southeast. Boundaries of these quadrants were marked on the edges of the pool with masking tape and labelled: North, South, East and West. A Plexiglas cylinder (13.75 cm x 9 cm diameter) was used as the escape platform in the maze. The cylinder was been filled with cement to weigh it down in the pool. The platform had a removable red and yellow striped top (3 cm x 9 cm in diameter) with a colourful flag erected in the center. For visible platform tests the level of the water in the pool was adjusted to 0.5-cm below the surface of the striped top, thus creating a visible escape platform, or to 0.5-cm above the white cylinder (with the striped top removed), creating a hidden escape platform.

The pool was located in a room measuring 5.2 x 2.4 m. Several posters were placed on the walls of the room to act as visual cues. There was also furniture in the room (sink, table, chairs) that provides visual cues. During testing, the room was dimly lit with diffuse white light (30 lux). The performance of the animals in the water maze was recorded using a video camera-based computer tracking system (Water maze, Actimetrics) on an IBM PC computer, with the camera fixed to the ceiling 2.1m above the pool.

In our paradigm, testing in the water maze takes 8 days: days 1-3 was acquisition training, days 4-6 reversal training, day 7 probe trial and day 8 was visible- platform day.

Acquisition and reversal training were done with the platform hidden (water is 0.5-cm above the escape platform). During reversal, the platform was moved to the opposite

side of the previous position. During the probe trial, there was no escape platform so that visuo-spatial memory can be assessed. On the visible-platform day the platform was moved to another quadrant of the pool and the visible top is added to the platform. This assesses basic visual ability and motivation to locate the platform. On each day, the mouse was removed from its home cage and was placed in a clean holding cage without woodchip bedding. Paper towel was torn into strips and placed in the bottom of the holding cages to allow the mice to dry more quickly. This paper towels were replaced when it became wet. Mice were run in squads of 4-6 with 5-minutes between each trial (inter-trial interval) for each mouse [29].

During acquisition training, the platform was placed in the center of the Northeast quadrant. Each mouse receives 4 trials per day. In each trial, the mouse was given a maximum of 60 seconds to locate the escape platform. The starting positions of the mice are predetermined using a Latin square design, which prevents the repetition of starting location sequences on back-to-back test days. Possible start positions were at the boundaries of the quadrants (e.g, West, North, East or South). For each trial, each mouse was removed from its holding cage using a small, clean 500-mL plastic container to minimize handling stress. The animal is then placed into the water at the appropriate start position. The mouse was then permitted to explore the pool and to search for the hidden escape platform for 60 seconds. When the animal located the platform, the timer was stopped (manually) and the mouse was allowed to stay on the platform. Once on the platform, the mice were permitted to view the extra-maze environment for 10 seconds. If the mouse failed to locate the platform during the allotted time, the animal was guided onto the platform using the plastic container. The next mouse is then placed in the pool and the same procedure followed. Each animal completes 4 trials per day over 3 days, for 12 trials of acquisition training, each trial from a different one of the 4 start locations. Reversal training begins on day 4. The invisible platform was moved to the opposite quadrant (Southwest quadrant), and mice are again assigned to appropriate start positions. The same procedures as in acquisition training were carried out during reversal training. Each of the animals completes 4 trials per day for 3 days for a total of 12 trials of reversal training.

A probe trial was conducted on day 7 to assess visuo-spatial memory. At this time, there was no escape platform in the maze. Each mouse was placed in the pool from one of the four possible start positions and allowed to explore the pool for 60-seconds, during which the time spent in each quadrant of the maze is recorded. When the 60-seconds are complete the mouse is scooped up using the container and placed in a holding cage to dry before being returned to its home cage.

The visible platform task was conducted on day 8. The visible platform was placed in a new location within the Northwest quadrant of the pool. The same procedures as in acquisition and reversal training are carried out and mice complete 4 trials.

During acquisition, reversal and visual training, the following behaviors were measured: swim latency (time to find and mount the escape platform), swim distance, proximity to the platform.

During the probe trial, the measures recorded are: frequency of entries into each quadrant (Northeast, Northwest, Southeast and Southwest), duration of time spent in

each quadrant, the number of times the mouse crosses the location of the platform during reversal training (annulus reversal crossing), the number of times the mouse crosses the location of the platform during acquisition training (annulus acquisition crossing), the duration and frequency of thigmotaxic behaviour (9 cm corridor width) and proximity to the platform location.

Beam walking for determination of motor coordination: motor coordination was assessed using the beam walking test. The beam walking apparatus was used to test motor coordination and balance. The beam walking test is more sensitive than the mouse rotarod in determining motor coordination deficits [34]. The beam has a length of 100 cm, a width of 2 cm and is elevated to a height of 40 cm. The beam is marked at 5 cm and 1 cm intervals. It is composed of wood and is coated with black paint. The animals were carried to the test room in their home cage. The mouse was removed from its home cage and placed at one end of the balance beam. After the mouse had secured its grip on the beam, the trial began. The maximum length of the trial is two minutes. The mouse was tested under white light, during the dark phase. The beam was cleaned with 70% ethanol and permitted to dry between each trial.

Behaviour scored were: Distance travelled, Foot Slips, Number of turns and Latency to fall.

Statistical Analysis

Data obtained were presented as mean \pm SEM. Experimental data were analyzed using analysis of variance (ANOVA) followed by a post hoc test (Least Square Difference (LSD) test) to determine significant difference between means. The analysis was done with an SPSS 18 statistical package. The mean values were considered significant at $p < 0.05$.

Results

Behaviours scored in morris water maze

Comparison of swim latency during the acquisition training: Figure 1 shows the swim latencies during the acquisition training of the Morris water maze (Days 1, 2 and 3) between the control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only. The swim latencies in day 1 were 59.95 ± 0.05 , 60.00 ± 0.00 , 59.97 ± 0.03 and 60.00 ± 0.05 seconds respectively. The result shown no significant difference between the experimental groups and the control in day 1. However, in day 2 the swim latencies were 49.11 ± 0.90 , 46.09 ± 0.00 , 47.43 ± 0.00 and 55.96 ± 0.04 seconds for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only respectively. There was a significantly longer latency for scopolamine only group compared to Control and other experimental groups ($p < 0.05$). The latencies were 33.14 ± 6.86 , 24.91 ± 5.01 , 28.69 ± 0.19 and 57.04 ± 0.82 seconds for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only respectively with the same trend like in day 2.

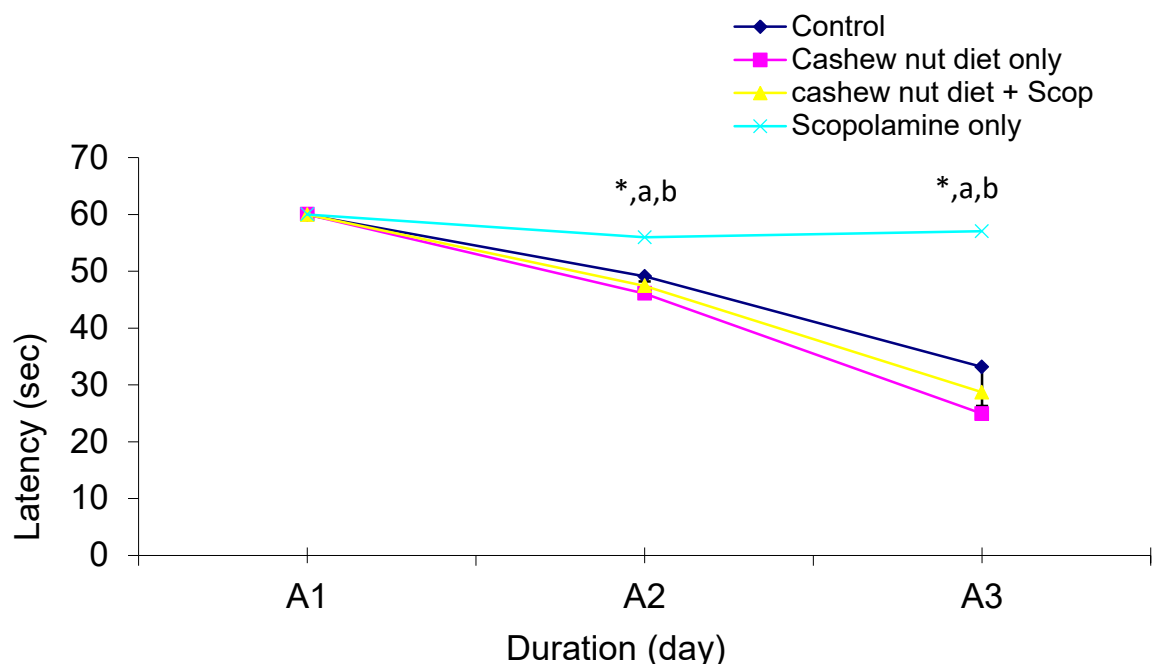


FIG. 1: Swim latency during acquisition training of the Morris water maze in control and test groups.

Values are expressed as mean +SEM, n = 7.

Comparison of swim latency during the reversal training: Figure 2 below shows the swim latency curves during the reversal training in the Morris water maze (Day 4, 5 and 6) for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only. The result shows that, in reversal training day 1 the swim latencies were 49.42 ± 0.69 , 51.60 ± 0.84 , 52.33 ± 1.60 and 57.45 ± 2.49 seconds respectively. In reversal training day two, the swim latencies obtained were 39.33 ± 2.20 , 41.05 ± 1.22 , 47.67 ± 0.43 and 60.00 ± 0.65 seconds for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only respectively. There was a significant difference between the scopolamine only test group compared to control and other experimental groups ($p < 0.05$). In the reversal training day 3, the swim latencies for control and the experimental groups were 26.45 ± 2.62 , 21.11 ± 2.81 , 31.71 ± 1.78 and 50.60 ± 0.43 seconds. There was a significant difference between the scopolamine only group compared to control and other experimental groups ($p < 0.05$). The result further shows that, the group fed cashew nut diet only shown a significant between the control and the group of mice fed cashew nut diet + scopolamine ($p < 0.05$).

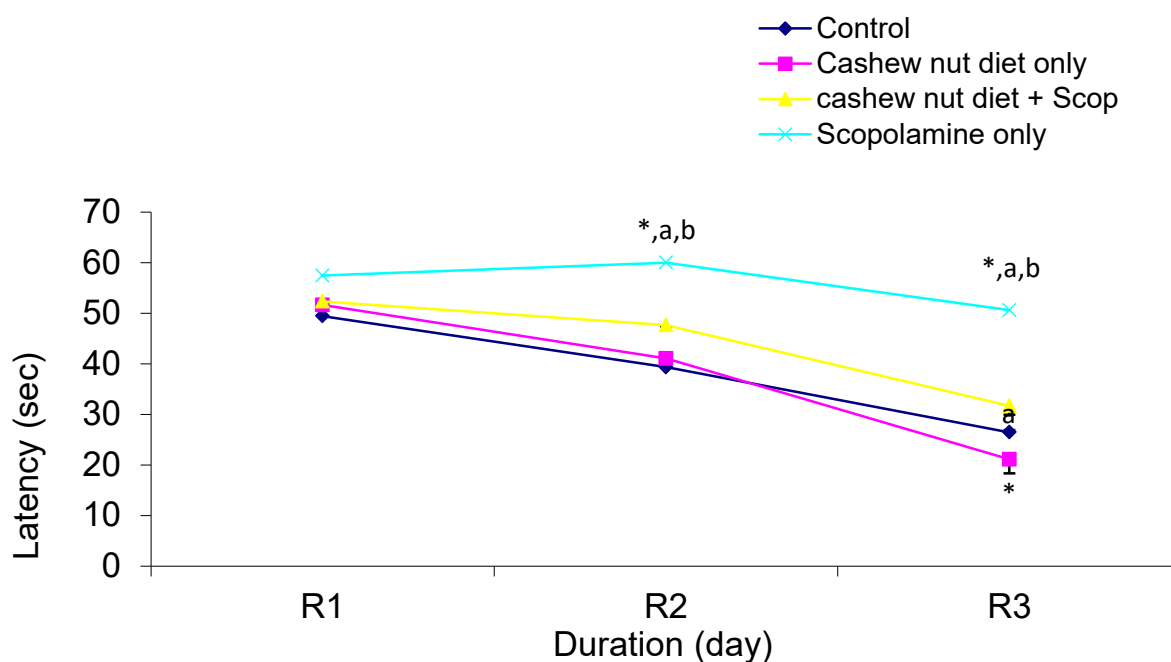


FIG. 2: Swim latency during reversal training of the Morris water maze in control and test groups.

Values are expressed as mean +SEM, n = 7.

* = $p < 0.05$ vs control

a = $p < 0.05$ vs cashew nut diet only

b = $p < 0.05$ vs cashew nut + scopolamine

Comparison of quadrant duration: Figure 3 below shows comparison of the quadrant duration in the Morris water maze test during the probe trial day. The mean durations in each quadrant for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only were northeast: 18.12 ± 1.27 , 15.97 ± 1.96 , 15.14 ± 0.00 , and 11.00 ± 0.01 , southeast: 11.45 ± 0.36 , 11.00 ± 3.00 , 13.94 ± 0.04 , and 18.46 ± 0.00 , southwest: 19.13 ± 0.65 , 21.44 ± 0.33 , 20.76 ± 0.28 , and 13.98 ± 0.02 and northwest: 11.30 ± 0.32 , 11.59 ± 1.24 , 10.16 ± 0.04 , and 15.22 ± 1.03 for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only respectively. The result shows that, the control group and the groups fed cashew nut diet show significant preference to the northeast and southwest quadrants compared to the group administered with scopolamine only ($p < 0.05$). The group fed cashew nut diet only shown the longest latency in the southwest quadrant.

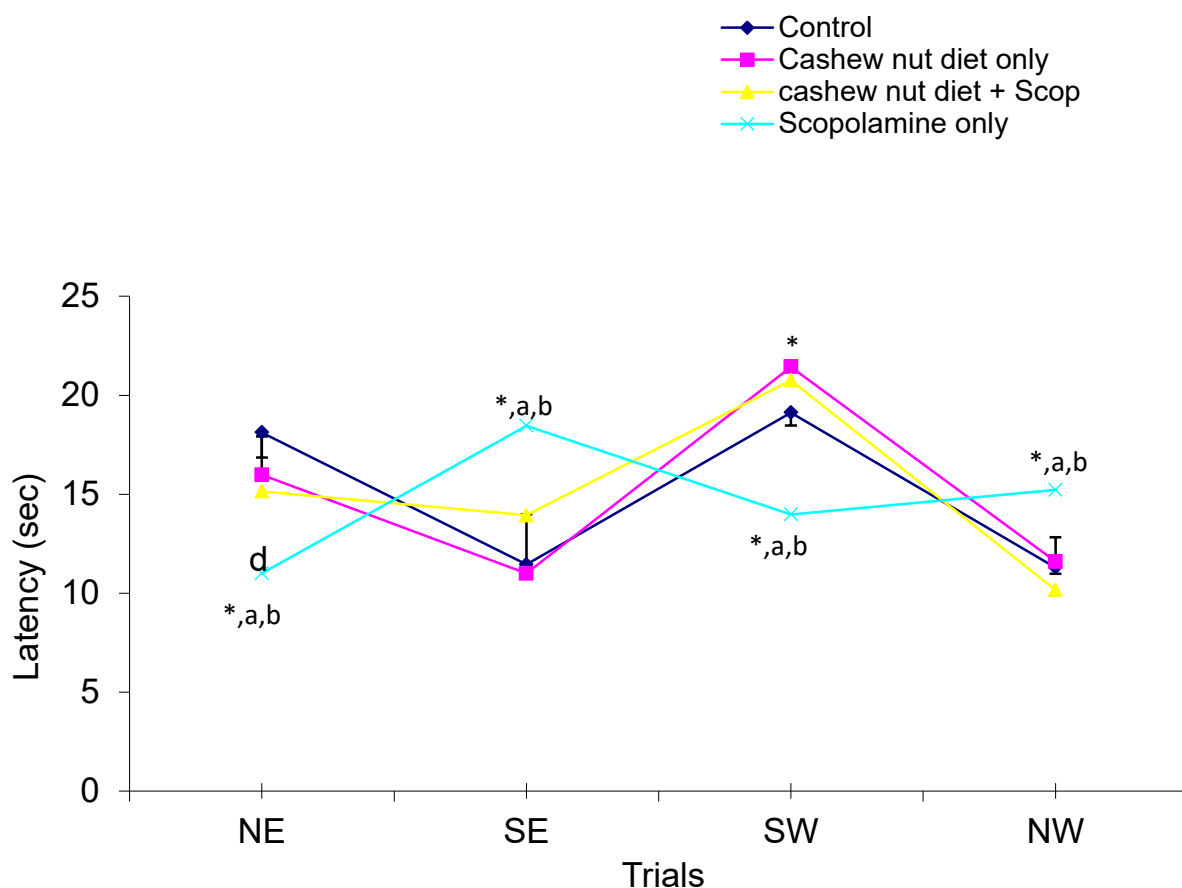


FIG. 3: Swim latency during probe trials of the Morris water maze in control and test groups.

Values are expressed as mean \pm SEM, $n = 7$.

* = $p < 0.05$ vs control

a = $p < 0.05$ vs cashew nut diet only

b = $p < 0.05$ vs cashew nut + scopolamine

Comparison of swim latency on the visible platform test: The swim latencies during visible platform test for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only were 9.30 ± 1.71 , 5.21 ± 0.93 , 8.41 ± 1.33 and 39.97 ± 1.32 . From the result, the swim latencies of the result the swim latency for the scopolamine only group was significantly longer compared to control and the groups fed cashew nut diets ($p < 0.05$). The group fed cashew nut only shown the shortest latency compared to the group fed cashew nut diet + scopolamine and the control group though there was no significant difference between them. The result is illustrated in figure 3 below:

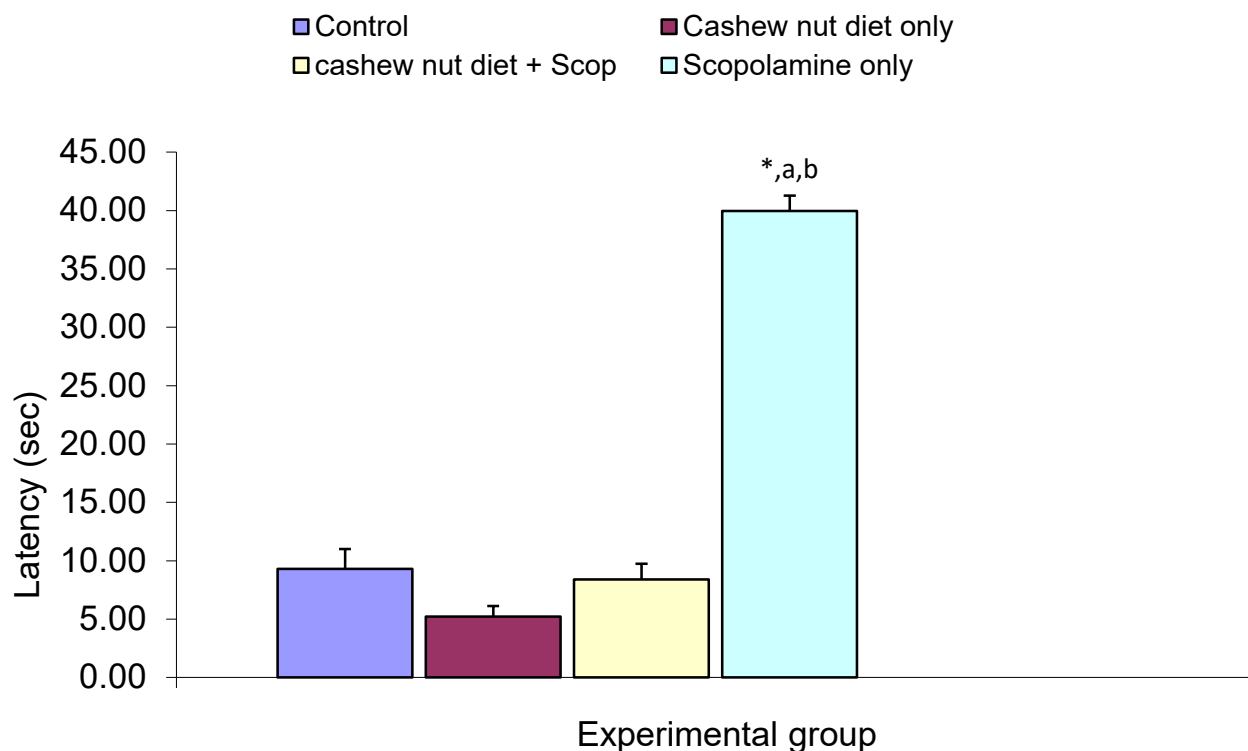


FIG. 4: Swim latency on the visible platform of the Morris water maze in control and test groups.

Values are expressed as mean +SEM, n = 5.

* = $p < 0.05$ vs control

a = $p < 0.05$ vs scopolamine

Behaviours scored in beam walking

Line crosses: Figure 5 below shows the comparison of mean line crosses for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only in beam walking apparatus as 302.50 ± 8.00 , 477.00 ± 3.34 , 426.25 ± 7.99 and 228.75 ± 9.08 respectively. The line crosses of the scopolamine only treated group shown significant decreased compared to control and other experimental groups ($p < 0.05$). The result further shown a significant difference between the groups fed cashew nut diets compared to control ($p < 0.05$). There was no significant difference between the group fed cashew nut diets.

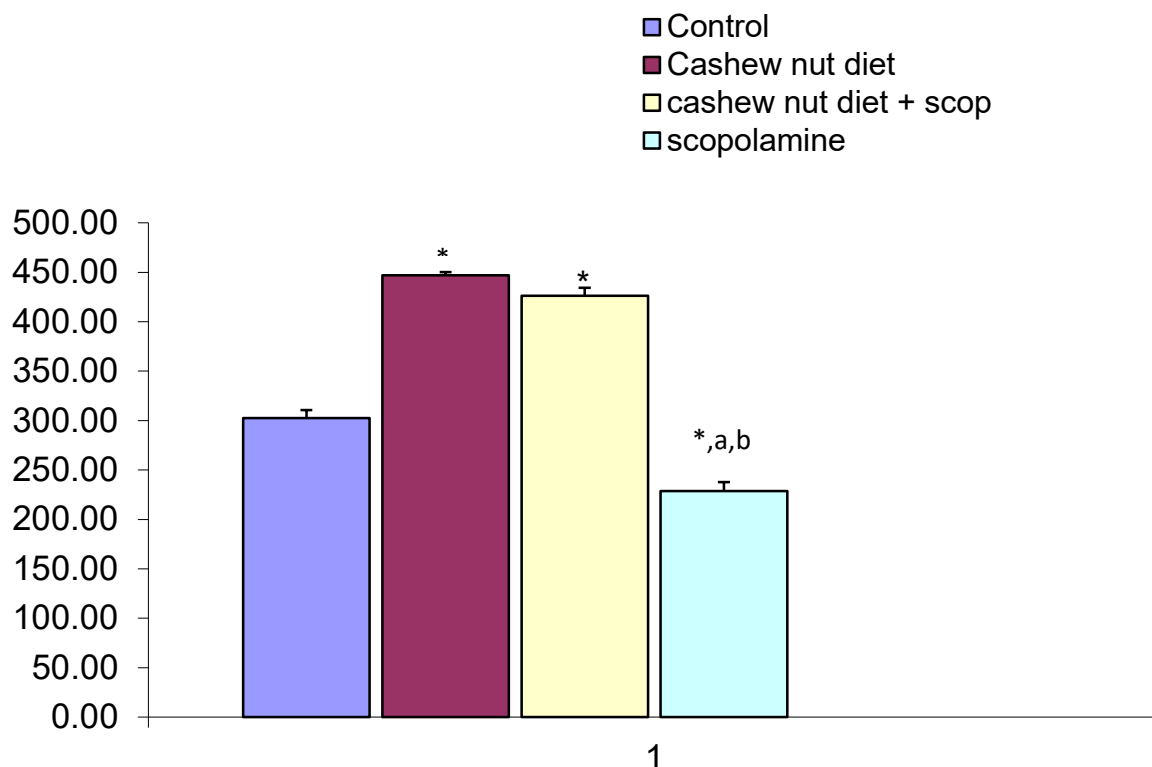


FIG. 5: Frequency of line crossing during the beam walking test in different experimental groups.

Values are expressed as mean ± SEM, n = 7.

* = $p < 0.05$ vs control

a = 0.05 vs cashew nut diet

b = $p < 0.05$ vs cashew nut + scop

Reversals: Figure 6 below shows the comparison of mean reversals of control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only in beam walking apparatus as 6.00 ± 0.08 , 5.80 ± 0.03 , 6.50 ± 0.06 and 3.50 ± 0.09 respectively. The results showed that, there was a significant decreased in mean number of reversals made by the scopolamine only treated group compared to control and the groups fed cashew nut diets ($p < 0.05$). There was no significant difference between the groups fed cashew nut diets compared to control.

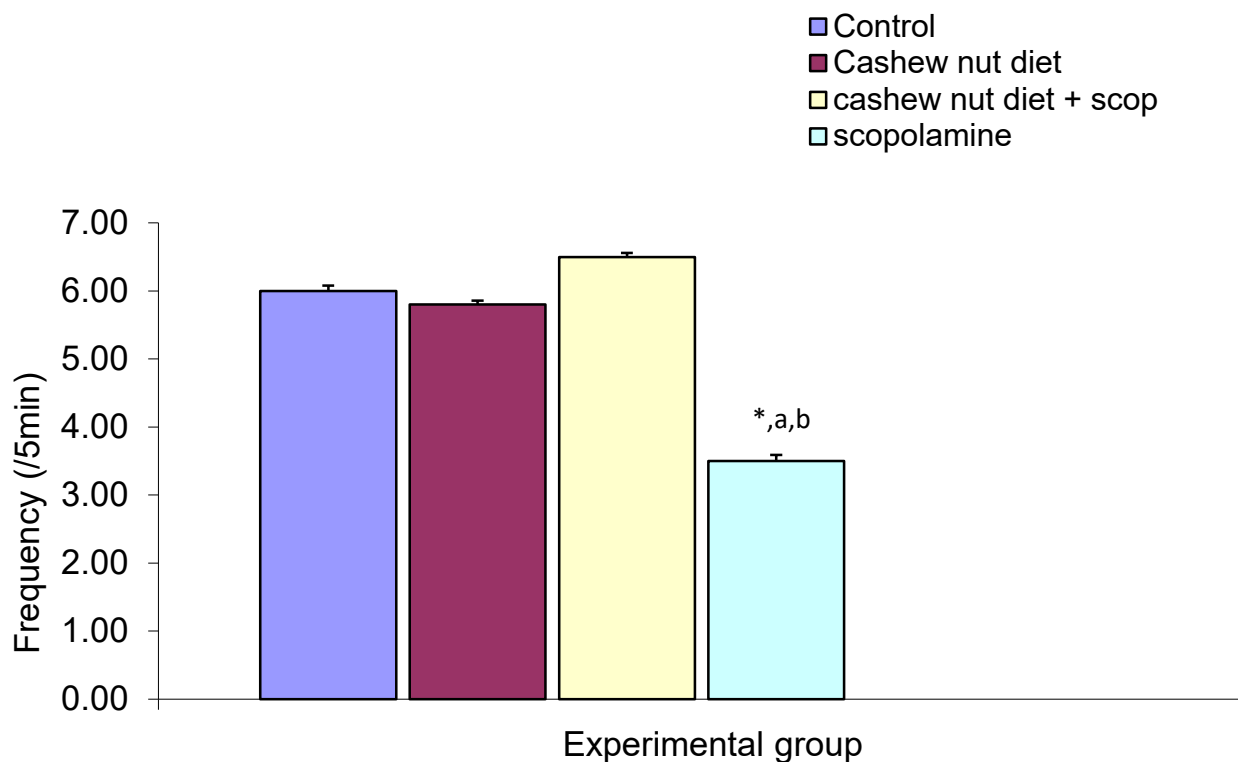


FIG. 6: Frequency of reversal during beam walk test in the control and test groups.

Values are expressed as mean +SEM, n = 7.

* = $p < 0.05$ vs control

a = 0.05 vs cashew nut diet

b = $p < 0.05$ vs cashew nut + scop

Foot Slips: The mean frequency of foot slips of the different experimental groups were recorded as follows: 1.75 ± 0.85 , 0.40 ± 0.98 , 3.50 ± 1.21 and 11.50 ± 0.29 for control, cashew nut diet only, cashew nut diet + scopolamine and scopolamine only in beam walking apparatus respectively. The result indicated that, the number of foot slips in the group treated with scopolamine only was significantly increased compared to control and the groups fed cashew nut diet ($p < 0.05$). There was no significant difference between the control and the groups fed cashew nut diets. The result is represented in figure 7 below.

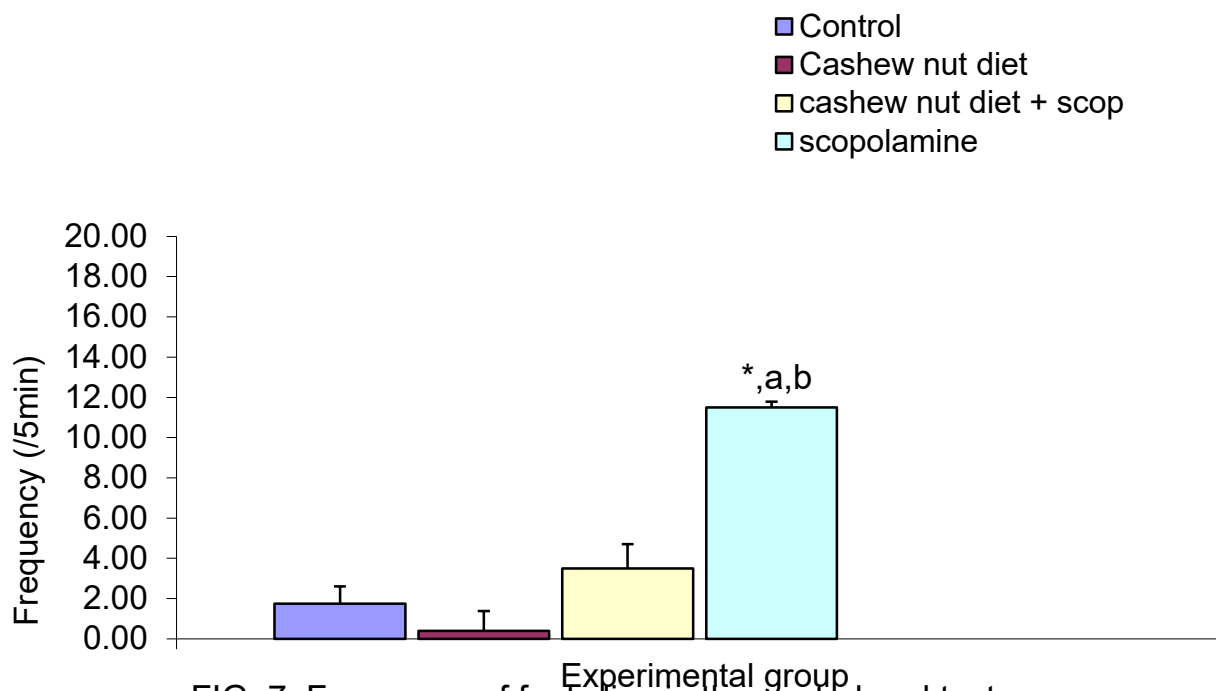


FIG. 7: Frequency of foot slips in the control and test groups during the beam walking test.

Values are expressed as mean +SEM, n = 7

* = $p < 0.05$ vs control

a = 0.05 vs cashew nut diet

b = $p < 0.05$ vs cashew nut + scop

Discussion

This study was to investigate the effect of consumption of cashew nut (*anacardium occidentale*) diet on scopolamine induced memory and motor coordination impairment in CD1 mice. The parameters considered in the study were learning and memory, motor coordination and balance. Morris water maze was used to study learning and memory whereas, the beam walking apparatus was also used to study motor coordination and balance. Before learning and motor coordination were studied, the administration of the test sample (cashew nut diet) and the placebo (control) was done for 21 days. The treatment period of 21 days was to enable sufficient time for bioavailability of nutrients, mainly protein, fat, vitamins, minerals and dietary fiber present in cashew nut. Recent research shows that cashew nut kernels are 43% rich in lipids and 20% rich in proteins and 37% rich in inorganic constituents. The inorganic components copper and iron act as cofactors for many physiological and metabolic functions [35]. Hence transient time is needed for biogenesis of serotonin from its precursor (tryptophan) present in cashew nut to be available in the brain tissues.

Learning and memory: Following the consumption of cashew nut diet and administration of scopolamine, the swim latencies for the first three days of acquisition training is illustrated in figure 1 above. The result shown that, the group administered scopolamine only had longer swim latency compared to control and the groups fed cashew nut diets ($p < 0.05$). There was no significant difference between the group fed cashew nut only compared to the group fed cashew nut + scopolamine. The same trend was observed during the reversal training days illustrated in figure 2 above. These mean that these groups of mice fed cashew nut diet were able to locate the hidden escape platform faster and so, learned faster compared to control group and scopolamine only treated group that consumed normal rodent chow though the result showed no significant difference compared to control. The group of mice administered scopolamine and normal rodent chaw had poorer learning curve during the acquisition and reversal training days. The acquisition and reversal test were used to assess the learning abilities of the respective groups of the test models. The day 7 test in the Morris water maze task is called probe trial used to assess long and short term memory represented in figure 3 above. The result showed that, the group of mice fed cashew nut diet only during probe test had longer duration spent in the northeast quadrant as well as in the southwest quadrant. The result further revealed the group fed cashew nut diet + scopolamine spending longer duration in both northeast and southwest quadrants, but with much longer duration in the southwest quadrant compared to northwest quadrant. The group of mice administered scopolamine only had no preference to any of the quadrants compared to the control and the groups fed cashew nut diet. The day 8 test in the morris water maze was used to assess visuospatial memory in the respective test groups. The result shown that, the group of mice administered scopolamine + normal rodent chaw had longer latency compared to control and other experimental test groups as illustrated in figure 4 above. The group fed cashew nut only had the shortest latency compared to control and the group fed cashew nut diet + scopolamine.

The cued version of the Morris water maze assesses cued learning and memory as well as visual integrity of the animal test models [36]. Impairments in performance in the hidden platform model may be due to some brain lesions or drugs which may affect the motivation to escape, or sensory motor factors rather than spatial learning [37]. This cueing procedure, in which the escape platform protrudes above the water surface, provides a control for this [38]. Here, the swim latencies were also used for the comparisons. Shorter swim latencies in the visible platform task indicate improved cued learning and longer swim latencies indicate poor cued learning. The probe trial in which the escape platform is absent was utilized to appraise hippocampus-dependent spatial long and short term memory. The mice administered scopolamine only had longer latency and shows no preference to any quadrant during the probe trial test as earlier explained. Recent researches had it that scopolamine is known to be a nonselective muscarinic receptor blocker. Doguc *et al.*, (2012) reported that, 0.8 and 2 mg/kg scopolamine administration significantly decreased glutamate ionotropic receptor N-methyl-D-aspartate (NMDA) type subunit 2A protein expression, which corroborates suggestions of an interaction between cholinergic and glutamatergic receptors in the hippocampus [39]. Whereas the group of mice fed cashew nut diet only

and the group fed cashew nut diet + scopolamine had shorter latencies and showed preference to the quadrants that contained the escape platform during the probe trail test. This shows that, the groups fed cashew nut diet expressed improved learning and memory. Which could be attributed to the fact that, cashew nut contained cholinergic and glutamatergic receptor agonists. Cashew nut contains vitamin B6, vitamin D, tryptophan and DHA that has a significant role in memory consolidation. Vitamin B6, comprising three chemically distinct compounds, pyridoxal, pyridoxamine, and pyridoxine, could be involved in the regulation of this improved mental function and mood [40]. The tryptophan present in cashew nut is the precursor of serotonin. The serotonergic system is a major contribution of consolidation of memory in the hippocampus projections for potentiation. It is suspected that, vitamin B6, D, tryptophan and DHA exert positive effects on NMDA receptors which are both ligand-gated and voltage-dependent involved in long-term potentiation, an activity-dependent increase in the efficiency of synaptic transmission for learning and memory [41].

Motor coordination: The results obtained from beam walking in this test showed that, the group(s) of mice fed cashew nut diet only and cashew nut diet + scopolamine showed better motor coordination compared to scopolamine only treated group. In figure 5 above representing the number of line crosses. The group of mice fed cashew nut diet only and the group fed cashew nut diet + scopolamine showed significantly higher number of line crosses compared to the groups administered scopolamine only. It is possible that scopolamine suppressed motor centers responsible for pattern generation in the brain as indicated in the results showing decreased number of line crossing. Scopolamine could also be the source of imbalance in gravitational environments and clumsy motor acts as indicated in figure 7 showing higher number of foot slips in the group of mice administered scopolamine only. These effects of scopolamine could be explained by its action on cholinergic muscarinic receptors in the vestibular system [42]. From the results obtained above, it could be suggestive that, cashew nut contains some neuroactive nutraceuticals that may have exerted effects on the cerebellum as well the basal ganglia. The result of this research corroborated Thach (1998) who suggested that, the cerebellum might indeed participate in both motor control and cognition, and in motor adaptation, motor learning, and procedural learning [43]. The result obtained within the group of mice fed cashew nut diet + scopolamine showed a significant difference with the group administered scopolamine only whereas, comparison with the group fed cashew nut only showed no significant difference. Motor incoordination exhibited in animals model induced with scopolamine reveals that, functional brain networks were impaired after the administration of a cholinergic receptor antagonist (scopolamine) in healthy subjects decreasing functional connectivity between several cortical regions in alpha, beta and gamma bands. In the alpha band, this reduction involved the left fronto-parietal and the inter-hemispheric fronto-temporal and parieto-occipital connectivity [44]. Therefore cashew nut diet is elucidating suspected effects on the cholinergic receptor of pattern generators in the cortex, basal ganglia, brain stem, and the anterior motor neurons of the spinal cord. Cashew nut contains vitamin B6, vitamin D, tryptophan and DHA. The enhancing cholinergic agonist activities of cashew nut is suspected from the serotonin precursor

(tryptophan) because, serotonergic pathways are included in the RS pathways activated by the locomotor signals from higher centers of the brain [18]. According to Bajoet *al.*, (2015) Scopolamine, a muscarinic receptor antagonist, produces a blocking of the activity of the muscarinic acetylcholine receptor and the concomitant appearance of transient cognitive amnesia and electrophysiological changes, which resemble those observed in Alzheimer's Disease [44]. The DHA has a major role in enhancing motor skills and neuronal myelination improving motor skills.

The effects of consumption of cashew nut (*Anacardium Occidentale*) diet on scopolamine-impaired memory and motor co-ordination in CD1 mice were studied. Learning was impaired in animals treated with scopolamine only compared control and other test groups ($p < 0.05$). Memory was also impaired in the group of animals treated with scopolamine only compared to control and other experimental groups ($p < 0.05$). Motor coordination was impaired in animals treated with scopolamine only compared to control and other experimental groups ($p < 0.05$). Consumption of cashew nut diets improved learning and memory as well as motor coordination. From the results obtained from this research, it is suspected that cashew nut diet has cognitive and muscarinic effects on hippocampus, basal ganglia, cerebellum, prefrontal cortex as well as the somatosensory cortex.

Conclusion: Consumption of cashew nut diet improved learning and memory as well as motor coordination. The learning, memory and motor coordination enhancement observed may be attributed to serotonin precursor (tryptophan), docosahexaenoic acid, vitamin B6 and vitamin D present in cashew nut diet.

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