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Ameliorative Effects of the Lyophilized Aqueous Seed Extract of *Buchholzia coriacea* on Scopolamine-Induced Memory Impairment in Sprague-Dawley Rats

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ABSTRACT

This study investigated the protective effects of lyophilized aqueous seed extract of *Buchholzia coriacea* (*B. coriacea*) in enhancing memory and neuroprotection in scopolamine-induced learning and memory impairment using Morris water maze and Y maze. Thirty-five adult male Sprague Dawley were divided into five groups (n=7) according to extract/drug administered. 14 days administration of two different doses (100 and 200 mg/kg) of lyophilized extract of *B. coriacea* seeds, donepezil (5 mg/kg) concomitantly with scopolamine (1 mg/kg i.p) was carried out. Assessment of Morris water and Y- maze tests, oxidative stress markers and histology demonstration (H and E stain) were carried out during and after administration to evaluate the memory enhancing activity of the lyophilized extract of *B. coriacea* seeds on scopolamine-induced memory impairment. The two doses of lyophilized extract of *B. coriacea* seeds, and especially the 200 mg/kg significantly decreased escape latency and increased number of crossing for Morris water maze, while spontaneous alternation was also increased in the Y maze compared to scopolamine only treated group. The extract increased the activity of superoxide dismutase, reduced glutathione and catalase while decreasing malondialdehyde. The extract mitigated the histological evidence of neurodegeneration observed induced by the use of scopolamine. In conclusion, lyophilized aqueous seed extract of *B. coriacea* poses to be a promising therapeutic agent for the treatment of cognitive dysfunction in addition to its already established medicinal properties.

Key words: *Scopolamine, Buchholzia coriacea, Memory loss, Neurodegeneration, Oxidative stress*

INTRODUCTION

Cognitive impairment is associated with stress, ageing, and neurodegenerative diseases including Alzheimer's (Shenkin et al. 2014; Ruano et al. 2019). Dementia, associated with neurodegenerative disorders have been reported to affect about forty eight million people globally (Feigin et al. 2019; Launer 2019). Decline in cognitive function as observed in dementia have been strongly linked to deficit of acetylcholine in the brain possibly to degeneration of cholinergic neurons in the hippocampus (Gu et al. 2015; Oz et al. 2016).

Scopolamine (hyoscine hydrobromide), a muscarinic acetylcholine receptor antagonist, has been reported to induce learning and memory impairment through the cholinergic neuronal system similar to

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Alzheimer's disease (Hasegawa et al. 2016). Blockade of the muscarinic-acetylcholine receptor that temporarily blocks synaptic communication between the neurons have been highlighted as possible mechanism in the induction of cognitive impairment by the use of scopolamine (Yoon et al. 2018; Bali et al. 2019; Aoki et al. 2020). Recently, treatment of cognitive impairment associated with neurodegenerative disorders have focused towards exploring this mechanism, hence, enhancing residual cholinergic neurotransmission via the use of acetylcholinesterase inhibitors (Oz et al. 2016; Yoon et al. 2018). Single cognitive enhancing agent like Donepezil, as well as co-administration such as galantamine plus Donepezil have proven to be effective in improving cognitive function (Zhang et al. 2019; Zhao et al. 2019). However, side effects including sleep disturbance have been reported to be associated with donepezil. Administration of donepezil 12 h before sleep has been reported to alter sleep cycle as inhibition of acetylcholine by the therapy promotes wakefulness (Zant et al. 2016; Kazui et al. 2017). Therefore the need for effective therapy with little to no side effects is still fast rising.

Over the decades, herbal medicine have been on the fore-front of research towards ameliorating neuropsychiatric effects induced by neurodegenerative disorders, as well as chemotherapy used in its management (Ratheesh et al. 2017; Farzaei et al. 2018). Phytochemicals including phenols, saponins and flavonoids present in medicinal plants have greatly contributed to the breakthrough in research involving the use of medicinal plants as adjuvant for both chemotherapeutic and neurodegenerative disorder-induced neuropsychiatric effect (D'Onofrio et al. 2017). *Buchholzia coriacea*, belonging to the family Capparaceae, commonly called wonderful kola, Musk tree and elephant kola (Oghenesuvwe et al. 2015), and locally called 'Ndo' in Mende (Sierra Leone), 'Doe-fish' in Kru-basa (Liberia), 'Eson-bese' in Akan-asante (Ghana), 'Kola Pimente' in French, 'Owi' in Edo, 'Uke' or 'Okpokolo' in Igbo, 'Uwuro' and 'Aponmu' in Yoruba (Nigeria) have been reported as a useful medicinal plant (Oghenesuvwe et al. 2015). Different parts of the plant are traditionally used for medicinal purposes including treatment for headache, otitis, sinusitis, bronchitis, and skin itch. Topically, the leaves are used to relieve fever, the seeds for difficult child birth, management of diabetes melllitus, hypertension and cold (Adisa et al. 2011; Olaiya et al. 2013). The seeds have been scientifcally proven to have antihelminthic and antimicrobial properties (Izah et al. 2018), cytotoxicity evaluation (Ajaiyeoba et al. 2003), and antibacterial activity (Mbata et al. 2009). Presence of phytochemicals such as β-sitosterol, alkaloids, flavonoids and phenol in *Buchholzia coriacea* seeds and its ability to cross the blood brain barrier have been linked to its antioxidant activities (Sayeed et al. 2016; Abayomi et al. 2019). In addition, a few studies report its neuroprotective property by effective inhibition of acetylcholinesterase, butyrlcholinesterase and monoamine oxidase enzymatic activities (Onasanwo et al. 2016; Adefegha et al. 2018). However, there is dearth of data as to its effect on scopolamine induced cognitive impairment. Therefore, this study was carried out to investigate the ameliorative effect of the lyophilized aqueous seed extract of *Buchholzia coriacea* on scopolamine-induced memory impairment in Sprague Dawley rats.

MATERIALS AND METHODS

Ethical Approval

Ethical approval was obtained from the College of Medicine of the University of Lagos with number: CMUL/HREC/04/19/511

Plant Collection and Extract Preparation

The fresh seeds were purchased from Mararaba market located in Abuja, Federal Capital Territory of Nigeria and taken to a qualified taxonomist in the Department of Botany, University of Lagos, Akoka, Lagos state, Nigeria for authentication. The Voucher number, LUH 8022 was given, specimen was deposited at the herbarium of the Department of Botany, University of Lagos, Akoka, Lagos state, Nigeria. The seeds were peeled, grated into small pieces then shade dried for 2 weeks after which it was pulverized into coarse powder using a local grinding machine. Extraction was done via cold maceration. One kilogram of the seed powder was extracted in distilled water for 72 h with manual intermittent shaking at 2 h interval. The extract was then filtered using Whatman No. 1 filter paper. The filtrate was concentrated using rotary evaporator (Rota vapor R 210, Büchi, Switzerland) at 40°C and then lyophilized using a freeze dryer. The yield was determined and the extract stored in a refrigerator at 4°C in sample containers prior to use.

Table 1: Phytochemical screening of lyophilized seed extract of *B. coriacea*

Saponins mg/100 g	Alkaloids mg/100 g	Reducing sugar mg/100 g	Cardiac glycoside mg/100 g	Flavonoids mg/100 g	Tannins mg/100 g	Steroids mg/100 g	Terpenoids mg/100 g
35.87	29.38	34.17	31.13	31.06	20.61	23.99	21.58
35.35	29.73	34.45	31.62	30.52	20.39	23.46	21.25

Phytochemical Analysis

In this study, the preliminary screening involved detailed phytochemical screening of the aqueous extract. The different qualitative and quantitative chemical tests were performed in order to establish the chemical composition of the extract. The phyto-constituents determinations were carried out using standard methods as described (Veeresham et al. 1994; Evans 2009). The phytochemicals analysed were alkaloids, flavonoids, saponins, terpenoids, steroids and tannins.

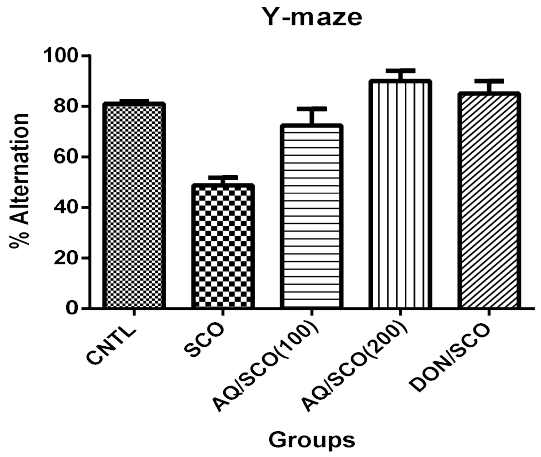

Figure 1: Percentile spontaneous alternation assessment of Y-maze test

Acute Toxicity Test

Five groups of rats (both sexes), each group containing five rats were used. Four groups were treated orally with varying doses of the *Buchholzia coriacea* seed extract at 250, 500, 1,000 and 2,000 mg respectively. Group 5 was given an equivalent volume of distilled water to serve as control. The animals were observed for toxic signs like excitability, dullness, diarrhoea and death over 72 h.

Experimental Animals

Male Sprague-Dawley rats with average weight of 180-220 g were used as test animals. They were obtained from the Laboratory Animal Unit of the College of Medicine, University of Lagos, Nigeria. The animals were housed in cages at room temperature at 12/12 h light/dark cycle for a period of two weeks prior to the commencement of the experiment. Standard commercial rat pellets and water were provided ad-libitum. The laboratory animals were used in accordance with laboratory practice regulation and the principle of humane laboratory animal care (Zimmermann 1983).

Drugs

Scopolamine hydrobromide (Sigma, USA) was used in this study. Scopolamine was dissolved in saline (NaCl 0.9%) at final concentrations of 1 mg/kg, and was injected intraperitoneally.

Experimental Design

A total of thirty-five Sprague Dawley rats were used. They were divided into five groups (n=7) according to the substances administered. Group 1: Positive control (distilled water only). Group 2: Negative control (scopolamine (1 mg/kg, i.p.) (Foyet et al. 2015). Group 3: Aqueous extract (100 mg/kg, orally, Ugwu et al. 2019) + Scopolamine (1 mg/kg) (i.p.). Group 4: Aqueous extract (200 mg/kg) orally (Ugwu et al. 2019) + Scopolamine (1 mg/kg) (i.p.). Group 5: Donepezil (5 mg/kg) orally (Li et al. 2018) + scopolamine (1 mg/kg) (i.p.).

All these groups received the corresponding treatment for 14 consecutive days. On each day, 30 min after the various treatments, scopolamine (1 mg/kg i.p.) was injected to all groups except the positive control group that received distilled water. The behavioural training and test began on day 10, 30 min after the injection of scopolamine.

Behavioural Assessment

Y-Maze Test

Y-maze test was used to evaluate short-term memory of the rats by recording spontaneous alternation in a single session on days 13 and 14. The maze used in this study was a Y-maze made of plywood with three identical arms (35 cm length × 8 cm height × 15 cm width) mounted at 120º to one another in a single piece. Each arm of the Y-maze was decorated with a different letter (A, B, or C) in order to be differentiated (Ma et al. 2007). One hour after the last treatment and 30 min after scopolamine injection (except for the distilled water group), each rat, previously naive to the maze, was placed at the end of one arm and were allow to move freely through the maze for five minutes. The number of arm entries was recorded for each rat. An arm entry was noted when a rat entered an arm of the maze with all its paws. Specific sequences of arm transitions (ABC, BCA, or CAB but not BAB or CAC or CBC) were recorded as spontaneous alternation that reflects short-term memory. The total number of arm entries reflects general locomotor activity. The arms of the maze were cleaned between sessions with 10% ethanol. The percentage of spontaneous alternation was defined according to the following equation: percentage of spontaneous alternation = [(Number of alternations)/(Total arm entries - 2)] × 100 (Ma et al. 2007; Kouémou et al. 2017). The effect on alternation behaviour was studied based on two parameters viz. percentage alternation and number of entries.

Morris Water Maze (MWM) Test

The MWM test was used to evaluate spatial long-term memory of rats. The MWM was performed as previously described by Morris (1984) with little modifications (Li et al. 2001; Kouémou et al. 2017). MWM consisted of a black circular pool (100 cm diameter, 50 cm height). The pool was located in a

room with various visual cues (pictures, shelters, curtains, lamps, fans). The position of the pool and that of the cues were maintained all the days of the experiment. The pool was filled with water at the temperature of 25 ± 2°C. The MWM was virtually divided into four equal quadrants: North, South, East, and West. A platform (11 cm diameter and 16 cm height) was centred in the South-East quadrant 1 cm below the water surface. The water was whitened by addition of liquid milk so that the platform was invisible at water surface. The position of the platform was kept unaltered during the training session. On the 1st day of the MWM test (familiarization, day 11 of drug treatment), 1 h after drug administration and 30 min after scopolamine injection, each rat received an acclimatization session where they were placed in the MWM for 60 s. During the acquisition phase (days 12 and 13 of drug treatment), 30 min after scopolamine injection, each rat was released into the pool, head facing the wall. The cutting time for each trial was 60 s. Rats that did not locate the platform within the time were gently guided to it and allowed to

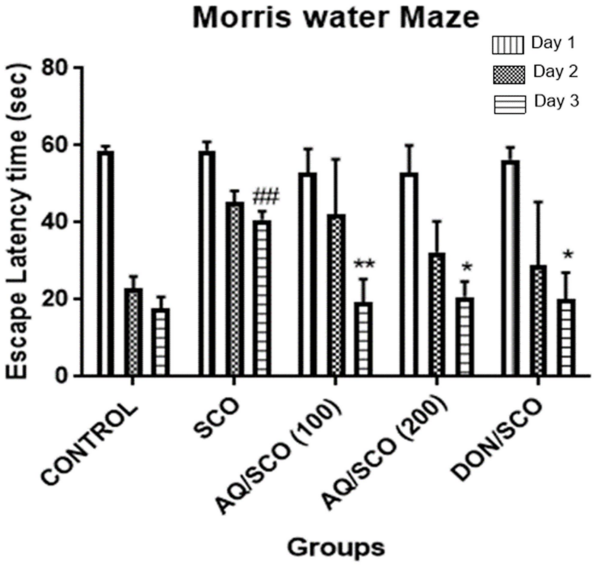

Figure 2: Escape latency time assessment of Morris water maze test. (###, p<0.001; ##, p<0.01; #: p<0.05 compared to control; *: p< 0.05, **: p<0.01, ***: p<0.001 compared to Scopolamine)

remain on it for 15 s. Each animal had three training sessions per day of 5 min interval. After each trial, each rat was taken to its cage and was allowed to dry up under a 60-watt bulb.

During each trial session, the time taken to reach the platform (escape latency) was recorded with stopwatches. In the retention phase (day 14 of drug treatment), the platform was removed from the pool (probe test). Each rat was placed into the MWM. The latency to reach the place of the former platform and the time spent in the target quadrant was recorded within 60 s using stopwatches.

Tissue Preparation

On day 14 following the MWM test, rats were decapitated under light chloroform anaesthesia. At the end of each sacrifice, the brain was immediately removed from the skull, rinsed and then weighed. Each brain was divided into two cerebral hemispheres; one half for biochemical assays, while the other half for histopathological analysis.

Biochemical Assays

Estimation of Protein Concentration

The total protein of brain homogenate was determined as described by Bradford (1976). 5 µL of the brain homogenate was introduced in microplate wells and 250 µL of Bradford reagent was added. After agitation, the absorbance of the mixture was read using a microplate reader at 590 nm (Kuo et al. 2013). The determination of the protein concentration was done using bovine serum albumin as standard.

Brain Reduced Glutathione Level

Reduced glutathione (GSH) level was estimated in the brain supernatant according to Ellman (1959). 20 µL of brain homogenates were mixed with 3 mL of Ellman reagent prepared in phosphate buffer (0.1 M, pH 7.2) at room temperature. After 1 h, the absorbance of the mixture was read at 412 nm. The amount of glutathione was calculated with the formula of Beer Lambert using the extinction coefficient value of 13,600/M/cm (Fotio et al. 2009). Each assay was done in triplicate.

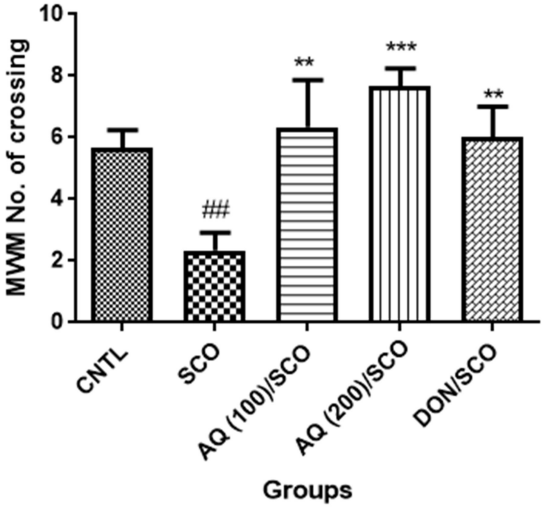

Figure 3: Line crossing assessment of Morris water maze test (###: p<0.01, ##: p<0.01, #: p<0.05 compared to control; *: p< 0.05, **: p<0.01, ***: p<0.001 compared to Scopolamine

Brain Malondialdehyde Level

The brain malondialdehyde (MDA) level was measured in the supernatant using the thiobarbituric assay. 1 mL of brain's supernatant was added to 0.5 mL of trichloroacetic acid (20%) and 1 mL of

thiobarbituric acid (0.67%). The mixture was heated in a water bath at 100°C for 60 min. After cooling, the mixture was centrifuged at 3000 rpm for 15 min. The absorbance of the supernatant was read at 530 nm. The amount of MDA was calculated with the formula of Beer Lambert using the extinction coefficient value of 1.56 × 105 M/cm. The concentration of MDA was expressed as µmol/g tissue (Kouémou et al. 2017). Each assay was done in triplicate.

Assay of Superoxide Dismutase Activity

Activity of superoxide dismutase in the brain was measured following Kakkar et al. (1984) with slight modifications. The reaction mixture was mixed vigorously with 4 mL of n-butanol and the mixture was allowed to stand for 10 min followed by centrifugation for 10 min at 3,000×g to separate the butanol layer. The colour intensity of the chromogen (purple) in butanol layer was measured at 560 nm against butanol on spectrophotometer. A mixture without enzyme preparations was run in parallel to serve as reagent blank. The activity of superoxide dismutase is expressed in units/min/mg protein. One unit of the enzyme is the amount required to inhibit the rate of chromogen formation by 50%.

Assay of Catalase Activity

The activity of catalase in the brain was assayed following the method of Aebi et al. (1984), using H2O2 as substrate. Briefly, reaction mixture in a final volume of 1.0 mL contained phosphate buffer (0.1 mM, pH 7.4), post mitochondrial fraction of sample (100 µL) and H2O2 (30 mM). The decrease in optical density was measured for 150 s at 240 nm using the spectrophotometer. The activity of the enzyme was calculated using the molar extinction coefficient 43.6 M/cm.

Histopathological Studies

After sacrifice, the brains were fixed in 10% formal saline. Fifty micrometre sagittal sections were made on the brain in the hippocampus region using the rat brain atlas with the following coordinates (anterior/posterior = -2.0 mm, medial/lateral = -1.5 mm, and dorsal/ventral. The brain sections were dehydrated in ascending concentrations of ethanol and followed clearing in xylene, then embedding in paraffin. Paraffin sections of the brain were de-paraffinized in xylene and rehydrated in descending concentrations of ethanol. Brain sections were then stained in haematoxylin and eosin. After drying overnight, the brain section images were captured using a digital camera attached to a light microscope.

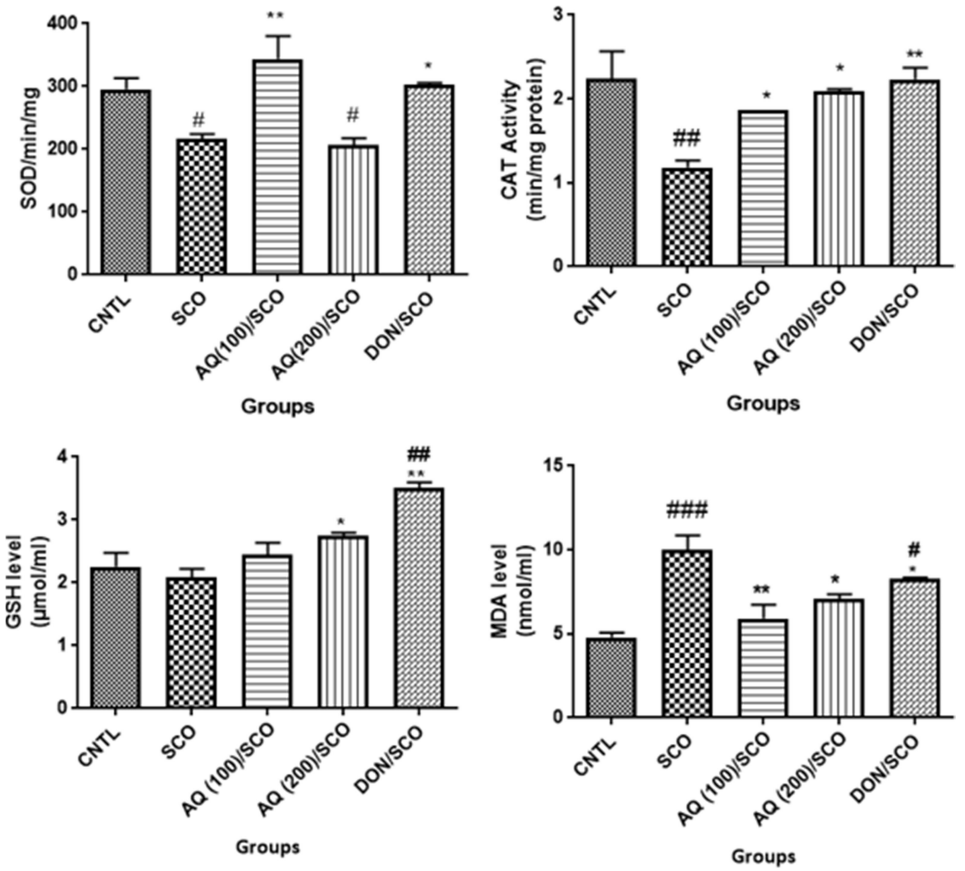

Figure 4: Oxidative stress marker assay (###: p<0.01, ##: p<0.01, #: p<0.05 compared to control; *: p< 0.05, **: p<0.01, ***: p<0.001 compared to Scopolamine; SOD: superoxide dismutase, CAT: catalase, GSH: reduced glutathione, MDA: malondialdehyde

Statistical Analysis

Statistical analysis was done using the software Graph-pad prism 6 for windows. The differences amongst groups were analysed using one-way analysis of variance. P - values ≤ 0.05 were considered significant.

RESULTS

Phytochemical Screening

Qualitative estimation revealed that aqueous seed extract of *B. coriacea* is rich in alkaloids, tannins, terpenoids, flavonoids, saponins, steroids, reducing sugars and cardiac glycoside. The detailed

summary of phytochemical screening of *B. coriacea* is shown in Table 1.

Acute Toxicity Study

No mortality was observed following oral administration of the aqueous seed extract of *B. coriacea* even at a dose of 2,000 mg/kg. More so, no significant change in body weight was observed. Hence, oral *B. coriacea* could be safe up to the dose of 2,000 mg/kg body weight of the animal.

mg/kg of *B. coriacea* treated groups showed no statistically significant change in escape latency time compared to control group. On day 3, only scopolamine treated group showed a statistically significant increase in escape latency time as compared to control group. Similarly, all treatment groups showed a statistically significant decrease in escape latency compared to scopolamine alone treated group (Fig. 2).

Line crossing assessment in the MVM test (probe test) also demonstrated that scopolamine treated

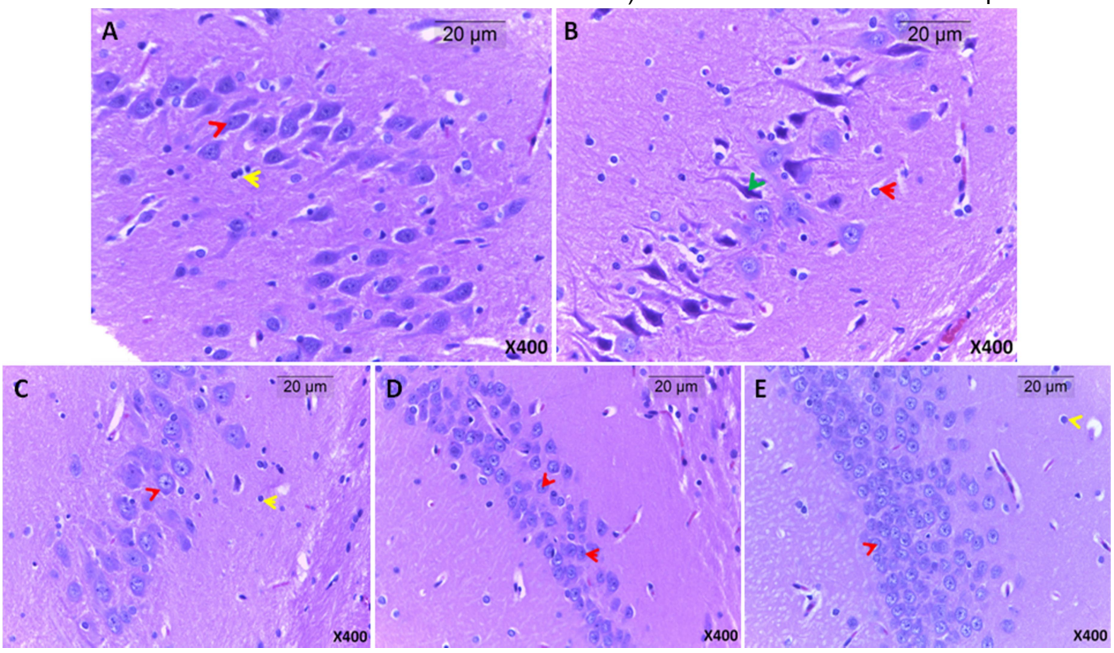

Figure 5: Photomicrograph showing histopathological changes in the CA3 of hippocampus in different treated groups (H&E ×400). a. Control group showing normal pyramidal cells. b. Scopolamine treated group showing marked degeneration of pyramidal cells (green arrow); c. DON (100 mg/kg) + scopolamine showing normal pyramidal cells (red arrow); d. Aqueous extract (BC)100mg/kg + scopolamine showing preserved cyto-morphology with very mild vacuolated nuclei. e. Aqueous extract (BC) 200mg/kg + scopolamine showing preserved cyto-morphology.

Y Maze Assessment

Y-maze test shows a significant decrease in percentile spontaneous alternation in the scopolamine only group in relation to the control group, while the use of *B. coriacea* seed as adjuvant significantly increased percentile spontaneous alternation (Fig. 1). 200 mg/kg of *B. coriacea* showed better improvement in percentile spontaneous alternation in Y-maze test compared to 100 mg/kg of the adjuvant.

Morris Water Maze Assessment

There is no statistically significant difference in escape latency among the treatment groups in comparison with the control after the first day of MVM training (Fig. 2). On trial day 2, there was a significant increase in the escape latency time in Scopolamine and 100 mg/kg of *B. coriacea* treated groups compared to the control, while, donepezil and 200

group significantly reduced the number of times the line was crossed during the probe test when compared to the control group, while other groups showed no statistically significant difference. However, 100 mg/kg, 200 mg/kg of *B. coriacea* and donepezil statistically improved the number of times the lines crossed during MVM test compared to scopolamine group (Fig. 3).

Biochemical Assays

Superoxide dismutase (SOD) activity level shows a statistically significant decrease in scopolamine only group in comparison to the control group, however, 100 mg/kg of *B. coriacea* and donepezil shows an increase in activity though not statistically significant when compared with control. There is a statistically significant increase in SOD activity level in both 100 mg/kg of *B. coriacea* and donepezil in comparison to

scopolamine group (Fig. 4). The 200 mg/kg group had the lowest activity.

Similarly, there is a statistically significant decrease in catalase (CAT) activity in scopolamine group compared to control, while other treated groups show no statistically significant difference in CAT activity in comparison with control. CAT activities were statistically significantly increased in the use of 100 mg, 200 mg of *B. coriacea* and donepezil as adjuvant compared to scopolamine only treated group.

In addition, reduced glutathione (GSH) level showed no statistically significant difference in all treatment groups except donepezil treated group compared to control (Fig. 4).

Lipid peroxidation level assayed via assessment of MDA elucidated that scopolamine treated group statistically increased MDA level significantly as compared to control. However, the use of *B. coriacea* at 100 mg/kg, 200 mg/kg and donepezil significantly reduced the level of MDA compared to the Scopolamine group (Fig. 4).

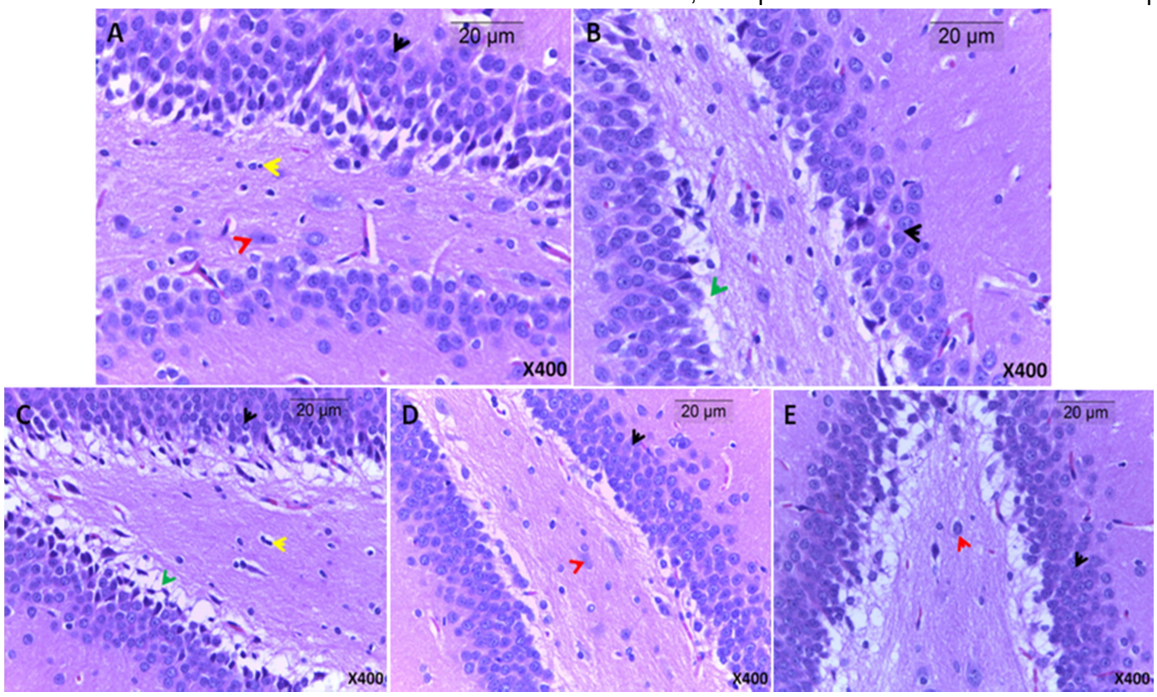

Figure 6: Photomicrograph of histopathological changes in the dentate gyrus in various treated groups (H&E, ×400). a. Control group showing normal granular cells. b. DON (100 mg/kg) + scopolamine showing normal granular cell layer but scanty towards the molecular layer (green arrow) c. Scopolamine treated group showing marked degeneration and vacuolation of granular cells (green arrow) d. Aqueous extract (BC) 100 mg/kg + scopolamine showing preserved cyto-morphology. e. Aqueous extract (BC) 200 mg/kg + scopolamine showing preserved cyto-morphology.

Histopathology

Large pyramidal shaped cells plus round to oval shaped nuclei of vesicular cells were observed in the cellular arrangement at the Cornu ammonis 3 (CA3) of the control group. Small rounded shaped nuclei of glial cells were also observed (Fig. 5a). The scopolamine-treated group showed changes in the form of shrinkage (neurodegeneration) of the pyramidal cells. Vacuolization of the cells (due to the degradation of the nuclei material), darkening of the nuclei (pyknosis) due to fragmentation and numerous rounded shaped nuclei of glial were also observed. However, donepezil group (100 mg/kg) with scopolamine showed marked reappearing of the cornu ammonis pyramidal cell layer, decrease in the neuronal loss and also less degeneration in the pyramidal cell layer of the hippocampus (Fig. 5e). *B. coriacea* seed extract (100 and 200 mg/kg) treated groups was found effective in restoring the alteration/ degeneration in the pyramidal cell layers and decreased the neuronal loss in CA3 region of the hippocampus which was affected by scopolamine. The higher dose (200 mg/kg) appears more effective in mitigating degeneration in the pyramidal cells when compared with scopolamine only treated group.

Similarly, normal cyto-morphology including proper alienation of granular cells along the limbs of the dentate gyrus was observed in the control group. However, multiple areas of vacuolization and atrophy of the granular cells at the medial surface of the limbs of the dentate gyrus were observed in scopolamine only treated group. Concomitant use of doses of extract of B. coriacea and donepezil (standard drug) proved to help preserve the cyto-architecture of the dentate gyrus with little to mild pathological changes induced by scopolamine (Fig. 6).

DISCUSSION

Dementia, a cognitive impairment, is one of the major health problems in normal aged life as well as in neurological disease conditions like Alzheimer's disease (Launer 2019). Cholinergic neurons present in the basal forebrain and hippocampus, with acetylcholine as a major neurotransmitter, are known to play vital roles in learning and memory processes (Ruano et al. 2019). Age-related dementia and memory deficit observed in AD have strongly been linked with the loss of cholinergic neurons in the basal forebrain and hippocampus (Gu et al. 2015; Oz et al. 2016). In addition, pharmacological blockage of cholinergic neurons in these areas causes impairment of memory and learning in experimental animals (Kouémou et al. 2017; Yoon et al. 2018). Therefore, findings of this study contribute to the existing knowledge on influence of hippocampal cholinergic neuron blockage on cognitive function and the use of 100 and 200 mg/kg of *B. coriacea* as a protective adjuvant.

Positive neuroplasticity activities at different cognitive centres of the brain especially at the hippocampus are strongly involved with memory formation and its consolidation (Vance et al. 2010; Budde et al. 2016). Kargbo (2017) reports that activity of acetylcholinesterase breaks down the acetylcholine release into the synaptic cleft resulting in negative neuroplasticity of new information learnt and its consolidation.

In this study, scopolamine reduced percentile spontaneous alternation in Y-maze test, increased latency time and decreased line crossing in MWM. This elaborates that scopolamine has a negative influence on cognitive neurobehavioral test via impairing learning and memory consolidation process in the experimental animals. This supports discussions that a non-selective muscarinic acetylcholine receptor antagonist like scopolamine may induce cognitive deficits and psychosis (Kargbo 2017; Iihara et al. 2019; Aoki et al. 2020). On the contrary, the use of *B. coriacea* improved cognitive function during neurobehavioral test possibly through inhibiting the activity of scopolamine on acetylcholinesterase in the synaptic cleft thereby significantly increasing the level of acetylcholine received by the post synaptic fibre. Similarly, Donepezil, a known cholinesterase inhibitor potentiates cognitive improvement against scopolamine. This corroborates the hypothesis and findings on activities of *B. coriacea* as an acetylcholinesterase, hence, accounting for its neuroprotective potentials as an anti-depressant (Onasanwo et al. 2016).

Incidences of increased oxidative stress have been strongly highlighted as a key mechanism of neurodegenerative disorders associated with dementia (Butterfield et al. 2017; Tarafdar et al. 2018). Over production of reactive oxygen species (ROS) is concurrent with imbalance in antioxidant enzyme activities. This agrees with hypothesis of studies that there is a strong link between scopolamine-induced dementia and increased oxidative stress (Haider et al. 2016; Balaban et al. 2017). However, the use of *B. coriacea* and donepezil improved antioxidant enzyme activities (increased SOD and CAT) and reduced ROS level (MDA) in this study. Phytochemical screening elucidated that the lyophilized aqueous seed extract of *B. coriacea* contains alkaloids, flavonoids, cardiac glycosides tannins and terpenoids. This correlates with phytochemical screening of both methanol and ethanol extracts of the plant (Ibrahim and Fagbohun 2013). Presence of these phytochemicals might have accounted for the improvement in scopolamine-induced increased oxidative stress. Plant extracts containing these phytochemical have been reported to reduce oxidative stress by either mopping up ROS, stimulation of antioxidant enzyme function or modulating inflammatory pathway against cellular death (Al-Dosari et al. 2017; Abayomi et al. 2019).

Incidence of lipid peroxidation as a result of increased MDA level is usually due to accumulation of unsaturated fatty acid present within the cellular membrane of the cells. This has been proven to trigger series of pathological changes including cellular deformity and disruption of structural and functional potency of cellular organelles (Ataie et al. 2016, Butterfield and Reed 2016; Abayomi et al. 2019). Histological assessment of this study demonstrated that administration of scopolamine induced pathological changes (vacuolization, pyknosis and cellular atrophy) to the pyramidal and granular cells of the hippocampus and dentate gyrus leading to cognitive impairment. This agrees with findings that scopolamine may have induced cognitive impairment as a result of indirect activities from increased oxidative stress and increased acetylcholinesterase activities than direct activity on the organelles of the hippocampus and prefrontal cortex (Wong-Guerra et al. 2017). Interestingly, *B. coriacea,* especially at 200 mg/kg optimally improved and ameliorated the pathological changes induced by scopolamine, similar to the ameliorating effect of donepezil. Restoration and preservation of cellular structure of the pyramidal cell of the hippocampus and granular cell of the dentate gyrus potentiates the effectiveness of *B. coriacea* in improving cognitive functions via direct action on the cellular structure or indirect action on antioxidant enzymes and accumulation of lipid peroxide within cellular membrane. This supports report that concurrent and reversal use of *B. coriacea* triggered marked regeneration, reduced vacuolation and pyknosis of neuronal cells of the prefrontal cortex (Abayomi et al. 2019).

Conclusion

The use of *B. coriacea* at 100 mg/kg, especially 200 mg/kg is shown to be effective in ameliorating cognitive impairment induced by scopolamine either directly by improving and preserving cellular structure of hippocampus pyramidal and granular cells of the dentate gyrus or indirect activity as antioxidants or acetylcholinesterase inhibitor, similar to mechanism by donepezil in improving cognitive function.

Conflict of Interest

None declared

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