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ARTICLE

Efficacy and Safety of Ashwagandha (*Withania somnifera* (L.) *Dunal*) Root Extract in Improving Memory and Cognitive Functions

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ABSTRACT

Objectives: Cognitive decline is often associated with the aging process. Ashwagandha (*Withania somnifera* (L.) *Dunal*) has long been used in the traditional Ayurvedic system of medicine to enhance memory and improve cognition. *Aim*: This pilot study was designed to evaluate the efficacy and safety of ashwagandha (*Withania somnifera* (L.) *Dunal*) in improving memory and cognitive functioning in adults with mild cognitive impairment (MCI). *Methods*: A prospective, randomized, double-blind, placebo-controlled study was conducted in 50 adults. Subjects were treated with either ashwagandha-root extract (300 mg twice daily) or placebo for eight weeks. *Results*: After eight weeks of study, the ashwagandha treatment group demonstrated significant improvements compared with the placebo group in both immediate and general memory, as evidenced by Wechsler Memory Scale III subtest scores for logical memory I ($p = 0.007$), verbal paired associates I ($p = 0.042$), faces I ($p = 0.020$), family pictures I ($p = 0.006$), logical memory II ($p = 0.006$), verbal paired associates II ($p = 0.031$), faces II ($p = 0.014$), and family pictures II ($p = 0.006$). The treatment group also demonstrated significantly greater improvement in executive function, sustained attention, and information-processing speed as indicated by scores on the Eriksen Flanker task ($p = 0.002$), Wisconsin Card Sort test ($p = 0.014$), Trail-Making test part A ($p = 0.006$), and the Mackworth Clock test ($p = 0.009$). *Conclusions*: Ashwagandha may be effective in enhancing both immediate and general memory in people with MCI as well as improving executive function, attention, and information processing speed.

KEYWORDS
ashwagandha; cognition; efficacy; memory; safety; *Withania somnifera* (L.) *Dunal*

Introduction

Cognition is the process by which the brain gathers, processes, and utilizes information. Memory is a critical component of cognition, as it allows information to be stored and retrieved for processing. Cognitive processes are involved in communication, reasoning, comprehension, problem-solving, decision-making, and judgment (Bostrom & Sandberg, 2009).

Cognition and memory tend to decline with increasing age. This decline may be associated with chronic somatic and neurodegenerative diseases; however, wide variability can be observed even among healthy individuals with respect to cognitive aging (Jelic, Kivipelto, &

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Winblad, 2006). A 10-year study recently found that cognitive decline often begins between the ages of 45 and 55 years (Singh-Manoux et al., 2012).

Mild cognitive impairment (MCI) is considered to be a transitional state between normal cognitive aging and dementia. It is characterized by memory problems without significant disruption in the activities of daily living (Chertkow, 2008).

Traditional systems of complementary medicine, such as Ayurveda and Traditional Chinese Medicine (TCM), have prescribed herbal therapies to aid memory and cognition for thousands of years. Evidence from clinical studies indicates that plant-derived therapies, such as *Bacopa monnieri, Ginkgo biloba, Panax ginseng, Salvia officinalis*, etc., may enhance memory and cognition and ameliorate symptoms of neurodegenerative disease (Aguiar & Borowski, 2013; Kaschel, 2009; Kennedy & Wightman, 2011; Miroddi et al., 2014).

Ashwagandha (*Withania somnifera* (L.) *Dunal*) is a staple therapeutic in the Indian Ayurvedic system of medicine, and is classified as a *Rasayana* or an "adaptogen." It also belongs to a sub-group of Rasayanas known as *Medhyarasayanas. Medhya* refers to the memory and intellectual capacity (Brahmachari, 2011; Department of Ayush, 1989; Singh & Udupa, 1993; World Health Organization (WHO), 2009). One of ashwagandha's prescribed applications is for treatment of memory and cognitive deficit following injury, illness, or simply old age (Singh & Udupa, 1993). Preclinical studies have indicated a potential role of ashwagandha as a nootropic, promoting cognitive function and enhancing memory (Dhuley, 2001; Naidu, Singh, & Kulkarni, 2006), presumably due to its cholinomimetic activity (Schliebs et al., 1997). Results have been promising for neurological disorders such traumatic brain injury, brain tumor, and neurodegenerative pathologies (Kuboyama, Tohda, & Komatsu, 2014; Singh, Narsimhamurthy, & Singh, 2008; Ven Murthy et al., 2010). Efficacy of ashwagandha extracts and isolated constituents has been reported in experimental models of neurodegenerative disorders such as Alzheimer's disease (AD) and Parkinson's disease (PD), which are associated with the disruption of neural networks and premature death of neurons (Kurapati et al., 2013; Nagashayana et al., 2000; RajaSankar et al., 2009; Sehgal et al., 2012). Clinical studies with ashwagandha have shown promise in bipolar disorder, enhancing cognitive and psychomotor performance, and ameliorating cognitive dysfunction (Chengappa et al., 2013; Pingali, Pilli, & Fatima, 2014).

To the best of our knowledge, however, the clinical impact of ashwagandha on the cognitive deficits seen in MCI has yet to be investigated. The aim of the present prospective, randomized, double-blind, placebo-controlled clinical pilot study, therefore, was to evaluate the efficacy and short-term safety of ashwagandha root extract in improving memory and cognition in human subjects with symptoms of MCI.

Materials and methods

This eight-week prospective clinical trial was conducted using a random-assignment, parallel-group, single-center, double-blind, placebo-controlled design. The protocol of the study was not modified after the trial's commencement.

Patient enrollment

Study subjects were selected from different outpatient clinics in the city of Pune, India, who sought treatment for MCI. Research coordinators invited the subjects to the study center at Chaitanya Hospital & Nursing Home, Pune, India for the study.

Inclusion in the present study required mild, subjective symptoms of memory impairment, age of 35 years or older, a previous diagnosis of early dementia or MCI or a score of ≥19 on the Mini-Mental State Examination (MMSE; Mungas, 1991), and the ability and willingness to provide informed consent. Exclusion criteria included a screening MMSE score of <19 (indicating moderate to severe memory impairment), known neuropsychiatric conditions, persistent endocrine disorders, uncontrolled hypertension or diabetes mellitus, drug dependence or addiction, use of alcohol, psychotropic drugs or drugs or alternative medicines for memory enhancement, or any severe comorbid medical condition. Pregnant or lactating women and subjects with known hypersensitivity to ashwagandha were also excluded from the study. Other than ashwagandha in the treatment group, the use of nootropic agents or anticholinesterase drugs was prohibited during the study.

Ethical consideration

The study was conducted in accordance with the Declaration of Helsinki (1989) and "Guidelines for Clinical Trials on Pharmaceutical Products in India – GCP Guidelines" issued by the Central Drugs Standard Control Organization, Ministry of Health, and Government of India. Institutional Review Board's approval was obtained from the study center at Chaitanya Hospital & Nursing Home, Pune, India. The ethical committee approval number was ECR/66/Inst/MH/2013. Ethics Committee notifications as per Good Clinical Practice Guidelines, issued by the Central Drugs Standard Control Organization and the Ethical Guidelines for Biomedical Research on Human Subjects, issued by Indian Council of Medical Research, were followed. Informed consent was obtained from each patient prior to study enrollment.

Study protocol and data collection

The study was initiated with a screening visit followed by an eight-week treatment period. At the screening visit, a brief medical history was obtained from each subject. A general physical examination was conducted, baseline body weight, BMI, and vital parameters were recorded, and the MMSE questionnaire (Mungas, 1991) was administered. A qualified psychiatrist then performed a clinical psychiatric examination on each subject to check for primary psychiatric disorders that would warrant exclusion from the study.

Investigational products

The investigational product, ashwagandha root extract (KSM-66 Ashwagandha) was received from Ixoreal Biomed (Los Angeles, CA, USA) as a gift sample (Batch #KSM/13/147). It is a 100% aqueous extract of *Withania somnifera* roots, containing 5% withanolides (determined by high performance liquid chromatography) as per the information provided by the manufacturer. This product is registered in India as traditional herbal medicine. The placebo capsules contained starch as inert filler. Placebo capsules were kept with a cloth-covered envelope containing ashwagandha root extract so that the odor of ashwagandha permeated to the placebo capsules.

Randomization and blinding

Following screening, research coordinators randomized the eligible subjects through a computer-based predetermined randomization (Rando version 1.0) in a 1:1 ratio to receive

either ashwagandha-root extract or placebo. The randomization list had non-stratified blocks of the same length. The study was a double-blind one, i.e., doctors and subjects were unaware of patients receiving the treatment and the placebo. Both drug and placebo were formulated as hard gelatin capsules having identical size, shape, color, texture, and weight. The treatment and placebo medication packs were tamper-proof and identical in appearance and weight. The packs were coded to conceal their contents, and the label contained the subject serial number (ID of the study). After the subjects were enrolled, they were provided with the medication pack having the corresponding serial number. During data collection, the research coordinators, the study investigators, and the attending care personnel were not allowed to access the randomization codes and allocations. Unblinding was allowed only after completion of the entire data collection process or in case of serious adverse events. The randomization codes were covered in aluminum foil and placed in a separate sealed envelope for each patient. Data analysts and persons in charge of reporting study results were also unaware of the identity of the study groups. The data were double-entered and blinded to the statisticians.

Interventions

The treatment group received 300 mg of ashwagandha root extract in capsule form, twice daily with water for eight weeks. The control group received an identical dose of placebo capsules. All subjects were evaluated at baseline and after four and eight weeks with the outcome measures described below. Data on safety and adverse effects were collected at the end of eight weeks. Patients Global Assessment of Tolerability to Therapy (PGATT) was also determined at eight weeks.

Outcome measures

The primary outcome measure for this study comprised a battery of cognitive tests assessing memory, visuospatial, executive function, and attention capabilities. These included the following instruments:

Memory

Wechsler Memory Scale III (WMS-III; Wechsler & Scale, 1997): The WMS-III is a validated, standardized instrument for the evaluation of several memory types. *Immediate memory* is the ability to remember a small amount of information over a few seconds. *General memory* is defined as the memory relating to personally experienced events that can be assessed experimentally with delayed recall of word lists, geometric designs, text, faces, and similar tasks. *Working memory* refers to the capacity to store information in the immediate term as the information is being received from perceptual inputs such as eyes, ears, and other sense organs. This capacity is needed to process and organize quickly arriving pieces of information for better assimilation and interpretation of information.

Owing to the locale of the present study, an adapted test called the Wechsler Memory Scale III, India (WMS-III INDIA; Wechsler & Gurappa, 2009) was used. This is a version of the WMS-III and has been adapted specifically for use in India.

As with the WMS-III, the WMS-III INDIA includes subtests for the primary indexes of immediate memory (subtests: logical memory I, verbal paired associates I, faces I, and family pictures I); general memory (subtests: logical memory II, verbal paired associates II, faces II, and family pictures II), and working memory (subtests: letter-number sequencing and spatial span).

Visuospatial processing and response

WMS-III INDIA Visual Reproduction I and II subtests: Assessment of recall, recognition, copy, and discrimination and recognition in the context of visual memory dysfunction. The subject is asked to recall and draw target images either while viewing them or from memory.

Shepard Mental Rotation Task (Cooper, 1975): Assessment of subject's cognitive rate of spatial processing. The subject is asked to compare images of two objects or letters, often rotated along a particular axis, and state whether the two are the same or are mirror images.

Executive function

Executive function is a higher order cognitive process relating to the coordination, selection, and execution of intentional action. This can involve the interruption of automatic but unproductive processes, or the reduction of impulses to stop a fruitful activity in which the body is engaged. Executive function was evaluated via the following tests:

Wisconsin Card Sort Test (Barcelo et al., 1997): Assessment of subject's abilities in abstract reasoning and the ability to shift cognitive strategies according to the demands of the situation. The subject is asked to match stimulus cards among a number of cards that differ with respect to color, quantity, and shape.

Eriksen Flanker Task (Eriksen & Eriksen, 1974): Assessment of subject's ability to focus on a central task and to suppress inappropriate responses. The subject is asked to identify the direction of a target image that is flanked by non-target stimuli.

Attention and speed of information processing

This cognitive component governs the extent of time for which the subject can receive information, filter out irrelevant information, focus on the relevant information, process that information, and respond accordingly. This component was assessed using the following tests:

Trail-Making Test Part A (Tombaugh, 2004): Assessment of subject's visual attention and task switching abilities; can provide information about visual search speed, speed of processing, and mental flexibility as well as executive functioning. The subject is instructed to connect a set of numbered dots in consecutive order as quickly as possible while still maintaining accuracy.

Mackworth Clock Test (Lichstein, Riedel, & Richman, 2000)*)*: Assessment of subject's vigilance and sustained attention capabilities. Over an extended period of time, subject is asked to note when a pointer on a round, clock-like background makes a double jump.

Safety and tolerability

All side effects and any adverse events were recorded for each subject. Patient Global Assessment of Tolerability to Therapy was then assessed on a 5-point scale: "excellent" (no adverse effects, and patient able to tolerate the drug), "good" (minimal side effects not interfering with patient's daily activities), "moderate" (some side effects and minimal interference in patient's daily activities), "poor" (significant side effects and significant interference in patient's daily activities), and "worst" (patient not able to tolerate the drug at all due to adverse effects).

Sample size determination

Based on standard deviation (SD) estimates of the outcome parameter scores from a small sample of four subjects, and a target minimal effect size of one on the outcome scores, the minimal required sample was calculated to be approximately 17 in each group. The sample size was increased to 25 per group to reduce inaccuracies in the SD estimates on this small sample size.

Statistical analysis

Quantitative data were expressed as mean score with SD; categorical and discrete data were expressed as numbers with percentages. Ranking data and scores were presented as mean score with SD. Confidence intervals (95% CI) were calculated wherever applicable. Since there was no exclusion of patients from the study or loss to follow-up, all analyses were done on per-protocol (PP) dataset using Med Calc Statistical Software, version 14.8.1. Demographic variables and vital parameters were compared between the treatment and placebo groups using one-way analysis of variance (ANOVA). Similarly, the two groups were compared for scores at all visits, and changes in scores were also analyzed using ANOVA. p-value was obtained using post hoc Mann–Whitney U test comparing the mean score between the treatment and placebo groups ($\alpha = 0.05$). Differences at the level of $p < 0.05$ were regarded as statistically significant.

Results

The recruitment and assessment of the subjects was performed from December 2013 to May 2014. A total of 50 adults over 35 years of age were enrolled in the study, and randomized to receive treatment with ashwagandha or placebo (Figure 1). At baseline, demographic parameters and cognitive impairment (as determined by MMSE) were similar in both groups (Table 1). Prior to treatment, subjective symptoms were also similar in both groups. Symptoms reported by subjects included: "Forgetting things more often" (100% of patients in both groups), "Forgetting important events such as appointments or social engagements" (88% in both groups), "Losing train of thought or the thread of conversations, books, or movies" (64% vs. 48%; $p = 0.254$), "Feeling increasingly overwhelmed by making decisions, planning steps

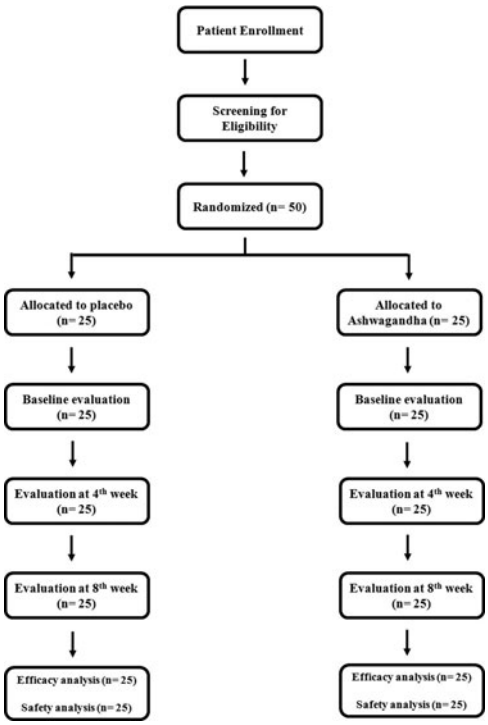

Figure 1. CONSORT flow diagram: Patient distribution and study design.

Table 1. Baseline characteristics.

	Ashwagandha ($n = 25$) Mean (SD)	Placebo ($n = 25$) Mean (SD)	t test p (t)
Age (year)	50 (7.33)	51 (7.98)	0.647 (0.213)
Weight (kg)	61.36 (6.8)	57.84 (6.34)	0.060 (3.711)
BMI (kg/m^2)	25.01 (2.68)	24.09 (2.64)	0.125 (2.284)
MMSE total score	23.2 (2.04)	24.56 (2.24)	0.093 (2.944)

to accomplish a task, or interpreting instructions" (32% vs. 40%; $p = 0.556$), and "starting to have trouble finding way around familiar environments" (20% in both groups). No significant differences were seen in vital parameters at baseline.

Mean baseline scores and change from baseline at four and eight weeks for the WMS-III INDIA are shown in Tables 2–4.

Improvement was observed in both treatment and placebo groups in the immediate memory subtest scores of WMS-III INDIA (Table 2). However, increased enhancement of scores for the ashwagandha treatment group versus placebo was statistically significant in the logical memory I ($p = 0.007$), verbal paired associates I ($p = 0.043$), faces I ($p = 0.020$), and family pictures I ($p = 0.006$) subtests after eight weeks.

Ashwagandha treatment also significantly enhanced scores compared with placebo in the general memory subtests of the WMS-III INDIA (Table 3). The logical memory II ($p = 0.006$), verbal paired associates II ($p = 0.031$), faces II ($p = 0.014$), and family pictures II ($p = 0.006$) subtests yielded significantly greater score enhancement for the treatment group compared with placebo after eight weeks. (The difference was not significant at four weeks ($p > 0.05$), apart from the family pictures II subtest ($p = 0.024$).)

Table 2. Wechsler Memory Scale (WMS-III INDIA) immediate memory subset: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Logical memory I	Mean score	SD	Mean score	SD	p
Baseline score	8.62	2.04	9.00	2.56	0.057
Score at 4 weeks (% change)	11.33 (31.4%)	2.51	10.42 (15.7%)	2.77	0.092
Score at 8 weeks (% change)	12.74 (47.8%)	2.69	11.04 (22.7%)	2.55	0.007
Verbal paired associates I	Mean score	SD	Mean score	SD	p
Baseline score	8.50	3.30	8.65	3.78	0.879
Score at 4 weeks (% change)	10.08 (18.6%)	2.69	9.03 (4.4%)	2.52	0.111
Score at 8 weeks (% change)	10.88 (28.0%)	2.53	9.69 12.0(%)	1.93	0.043
Faces I	Mean score	SD	Mean score	SD	p
Baseline score	10.38	2.78	10.73	2.93	0.662
Score at 4 weeks (% change)	12.76 (22.9%)	2.46	11.88 (10.7%)	2.52	0.090
Score at 8 weeks (% change)	13.88 (33.7%)	2.52	12.58 (17.2%)	2.33	0.020
Family pictures I	Mean score	SD	Mean score	SD	p
Baseline score	11.04	3.21	11.65	3.50	0.522
Score at 4 weeks (% change)	13.33 (20.7%)	2.10	12.46 (7.0%)	2.58	0.030
Score at 8 weeks (% change)	14.32 (26.4%)	1.79	13.00 (11.6%)	2.06	0.006

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean score between treatment and placebo groups (two-tailed $\alpha = 0.05$).

Table 3. Wechsler Memory Scale (WMS-III INDIA) general memory subset: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Logical memory II	Mean score	SD	Mean score	SD	p
Baseline score	7.50	2.73	7.73	2.86	0.772
Score at 4 weeks (% change)	10.29 (37.2%)	2.89	9.08 (17.4%)	2.77	0.078
Score at 8 weeks (% change)	11.38 (51.7%)	2.56	9.50 (22.9%)	2.58	0.006
Verbal paired associates II	Mean score	SD	Mean score	SD	p
Baseline score	9.21	3.16	8.65	2.67	0.508
Score at 4 weeks (% change)	10.21 (10.9%)	1.79	8.88 (2.7%)	1.95	0.152
Score at 8 weeks (% change)	10.79 (17.1%)	1.67	9.19 (6.2%)	1.65	0.031
Faces II	Mean score	SD	Mean score	SD	p
Baseline score	9.92	2.45	9.08	2.02	0.195
Score at 4 weeks (% change)	12.34 (24.3%)	2.00	10.73 (18.2%)	2.10	0.194
Score at 8 weeks (% change)	13.38 (34.9%)	1.84	11.23 (23.7%)	1.76	0.014
Family pictures II	Mean score	SD	Mean score	SD	p
Baseline score	11.29	2.82	11.19	2.73	0.900
Score at 4 weeks (% change)	13.67 (21.1%)	1.64	12.46 (11.3%)	1.71	0.024
Score at 8 weeks (% change)	14.46 (28.1%)	1.93	12.96 (15.8%)	1.42	0.006

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean scores between treatment and placebo groups (two-tailed $\alpha = 0.05$).

Results were inconclusive for the working memory index (Table 4). Treatment produced significant improvement in the spatial span subtest score ($p = 0.044$) after eight weeks compared with placebo but the effect on letter number sequencing subtest score was not significant ($p = 0.154$).

Mean baseline scores and mean change in baseline scores for the battery of cognitive function tests after four and eight weeks are presented in Tables 5.

Visuospatial processing and response (Table 5) were assessed with the visual reproduction I and II subtests of WMS-III INDIA as well as the Shepard mental rotation task. No statistically significant treatment effect was observed in these scores ($p > 0.05$).

Executive function was assessed with the Wisconsin card sort test and the Eriksen Flanker task (Table 6). Treatment with ashwagandha produced significant enhancement of test scores for both of these measures versus placebo at four weeks (Wisconsin card sort test, $p = 0.03$;

Table 4. Wechsler Memory Scale (WMS-III INDIA) working memory subset: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Letter number sequencing	Mean	SD	Mean	SD	p
Baseline	11.88	2.86	12.85	2.15	0.185
Score at 4 weeks (% change)	12.34 (%)	2.21	12.81 (%)	1.75	0.386
Score at 8 weeks (% change)	12.88 (%)	1.74	13.08 (%)	2.01	0.154
Spatial span	Mean	SD	Mean	SD	p
Baseline	12.12	3.31	11.65	3.54	0.629
Score at 4 weeks (% change)	12.83 (%)	2.37	11.42 (%)	1.90	0.131
Score at 8 weeks (% change)	1.71 (%)	1.69	11.69 (%)	1.59	0.044

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean scores between treatment and placebo groups (two-tailed $\alpha = 0.05$).

Table 5. Visuospatial processing and response: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Visual reproduction I (WMS-III INDIA)	Mean	SD	Mean	SD	p
Baseline	12.71	3.43	12.65	2.84	0.952
Score at 4 weeks (% change)	13.04 (2.6%)	2.04	12.73 (0.6%)	2.21	0.671
Score at 8 weeks (% change)	13.5 (6.2%)	1.84	12.73 (0.6%)	1.72	0.163
Visual reproduction II (WMS-III INDIA)	Mean	SD	Mean	SD	p
Baseline	11.83	3.07	11.69	2.84	0.867
Score at 4 weeks (% change)	12.41 (4.9%)	1.89	11.61 (–0.6%)	1.62	0.193
Score at 8 weeks (% change)	12.66 (7.0%)	1.46	11.77 (0.7%)	1.72	0.100
Shepard mental rotation task	Mean	SD	Mean	SD	p
Baseline	4.04	1.40	4.23	1.58	0.656
Score at 4 weeks (% change)	4.37 (8.2%)	1.46	4.69 (10.9%)	1.68	0.774
Score at 8 weeks (% change)	4.92 (21.8%)	1.54	4.65 (9.9%)	1.60	0.315

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean scores between treatment and placebo groups (two-tailed $\alpha = 0.05$).

Eriksen Flanker task, $p = 0.017$), which was further increased at eight weeks (Wisconsin card sort test, $p = 0.014$; Eriksen Flanker task, $p = 0.002$).

Attention and information processing speed were assessed through the trail-making test part A, and the Mackworth Clock test (Table 7). Significant enhancement in both scores was observed in the treatment group versus placebo after four weeks (trail-making test part A, $p = 0.039$; Mackworth Clock test, $p = 0.021$), and further enhancement was seen at eight weeks (trail-making test part A, $p = 0.006$; Mackworth Clock test, $p = 0.009$).

After treatment for four or eight weeks, the vital parameters (systolic and diastolic blood pressure, pulse rate, temperature, and respiratory rate) were similar for ashwagandha treatment and placebo (ANOVA; $p > 0.05$ for all). The study medication was very well tolerated by the study subjects. No adverse event was reported by any subject in either of the study arms. As per PGATT, all subjects in both group reported an "excellent" tolerability.

Discussion

This prospective, randomized, double-blind, placebo-controlled clinical trial was a pilot study to evaluate the efficacy of ashwagandha root extract in improving memory and cognition in 50

Table 6. Executive function: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Wisconsin card sort test	Mean	SD	Mean	SD	p
Baseline	48.92	22.19	51.23	16.40	0.679
Score at 4 weeks (% change)	42.50 (–13.1%)	5.94	18.69 (–5.0%)	6.30	0.030
Score at 8 weeks (% change)	40.96 (–16.2%)	5.24	47.08 (–8.1%)	5.29	0.014
Eriksen Flanker task	Mean	SD	Mean	SD	p
Baseline	5.62	2.12	5.65	2.26	0.963
Score at 4 weeks (% change)	6.12 (8.9%)	0.88	5.61 (–0.7%)	0.60	0.017
Score at 8 weeks (% change)	6.37 (13.3%)	0.79	5.73 (1.4%)	0.63	0.002

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean scores between treatment and placebo groups (two-tailed $\alpha = 0.05$).

Table 7. Attention and information processing speed: Mean scores and percentage change at four and eight weeks.

	Ashwagandha ($n = 25$)		Placebo ($n = 25$)		
Trail-making test part A (time required)	Mean	SD	Mean	SD	p
Baseline	68.17	21.59	62.19	26.70	0.387
Score at 4 weeks (% change)	63.25 (–7.2%)	4.56	59.69 (–4.0%)	3.30	0.039
Score at 8 weeks (% change)	61.79 (–9.3%)	4.15	58.73 (–5.6%)	2.66	0.006
Mackworth clock test	Mean	SD	Mean	SD	p
Baseline	8.88	2.03	9.12	1.68	0.652
Score at 4 weeks (% change)	9.55 (7.5%)	0.92	9.12 (0.0%)	1.06	0.021
Score at 8 weeks (% change)	9.76 (9.9%)	1.08	9.16 (0.4%)	1.08	0.009

p-value was obtained using the post hoc Mann–Whitney U test comparing the mean scores between treatment and placebo groups (two-tailed $\alpha = 0.05$).

human subjects with symptoms of MCI. A detailed analysis revealed that daily treatment with ashwagandha for a period of eight weeks produced significant enhancement versus placebo in a battery of cognitive tests designed to assess memory ($p \leq 0.05$), executive function ($p \leq 0.03$), and attention and information-processing speed ($p < 0.01$). Treatment effects on working memory and visuospatial processing assessments were inconclusive ($p > 0.05$).

Mild cognitive impairment is frequently a precursor of AD. MCI involves subjective complaints of memory loss and decline from normal function, with unchanged basic functioning; no medical, neurological, or psychiatric explanation for memory loss, and cognitive impairment not meeting the criteria for dementia (Chertkow, 2008). Owing to the presumed pathophysiological relationship between MCI and AD, the therapeutic approach to both conditions includes acetylcholinesterase inhibitors, nootropics, antioxidants, and anti-inflammatory drugs (Jelic & Winblad, 2003).

Ashwagandha root has been found to possess antioxidant, neuroprotective, anti-inflammatory, antidepressant, anxiolytic, and immunomodulating activities (Verma & Kumar, 2011). Plant-derived compounds from ashwagandha have shown significant potential as an acetylcholinesterase inhibitor (Choudhary et al., 2005, 2004; Vinutha et al., 2007), which would support its utility for the treatment of MCI and AD.

Working memory can be modulated by drugs having dopaminergic activity (Barch, 2004) and a number of studies have found that ashwagandha root can stimulate the dopaminergic system of the brain (Prakash et al., 2014; RajaSankar et al., 2009). A number of animal studies and clinical trials support the nootropic activity of ashwagandha (Chengappa et al., 2013; Dhuley, 2001; Naidu, Singh, & Kulkarni, 2006; Pingali, Pilli, & Fatima, 2014). Evidence indicating neuroprotective effects of ashwagandha supports its therapeutic efficacy in MCI and AD patients (Kuboyama, Tohda, & Komatsu, 2014; Singh, Narsimhamurthy, & Singh, 2008; Ven Murthy et al., 2010).

Ashwagandha also possesses anti-stress, anxiolytic, and sedative properties. These may be indirectly involved in improving memory and cognition in human subjects, as stress, anxiety, and sleep disorders can affect normal cognitive function (Eysenck et al., 2007; McEwen & Sapolsky, 1995; Miller, 2015). For example, highly stressed caregivers of terminally ill patients have been found to experience cognitive impairment relative to healthy normative subjects (Mackenzie et al., 2007). A study of fire fighters revealed an inverse relationship between stress and cognitive performance: As stress reactions increased, a reduction in controlled task focus was observed (Kivimäki & Lusa, 1994). Anxiety is another factor that may impair cognition by decreasing attentional control and increasing attention to threat-related stimuli (Eysenck et al., 2007).

In the present study, an adaptation of the WMS-III instrument (WMS-III INDIA) was used to assess the effect of ashwagandha on immediate, general, and working memory as well as visuospatial processing and response in subjects with MCI. The logical memory I and verbal paired associates I subtests of the WMS-III are useful for measuring auditory immediate response, which is mediated by the left posterior temporo-parietal cortex of the brain (Baldo, Katseff, & Dronkers, 2012). The logical memory II and verbal paired associates II subtests measure auditory declarative response, which is mediated by various parts of the brain, including the temporal and parietal lobes, prefrontal cortex, hippocampus, and amygdala. Treatment with ashwagandha was associated with significant enhancement in auditory immediate and declarative memory compared with placebo, as seen in the logical memory I and II and verbal paired associates I and II subtest scores after eight weeks ($p < 0.05$).

The faces I and family pictures I subtests of WMS-III INDIA were utilized to assess the visual immediate response, and the faces II and family pictures II subtests were used to assess the visual declarative response of the subjects. The visual immediate response is mediated through posterior parietal cortex (Todd & Marois, 2004) and the visual declarative response utilizes the posterior parietal cortex, occipital lobe, and dorsal and ventral stream pathways. In this study, treatment with ashwagandha was associated with significant enhancement in visual immediate and declarative memory compared with placebo after eight weeks ($p < 0.05$).

The letter number sequencing and spatial span subtests of WMS-III INDIA were utilized to assess working memory. Eight weeks of treatment with ashwagandha was associated with significant enhancement in the spatial span scores compared with placebo ($p < 0.05$); however, treatment did not significantly enhance letter number sequencing scores ($p > 0.05$). Working memory has multiple aspects, including working memory for spatial patterns and working memory for symbols. The results of this study indicate that ashwagandha may enhance working memory for spatial patterns but may have little impact on the working memory for symbols. Working memory is regulated through the prefrontal cortex of the brain. The dorsolateral areas of the prefrontal cortex are responsible for spatial working memory, while the ventrolateral areas are involved in non-spatial working memory (Owen, 1997). Ashwagandha may produce differential effects in those regions of the prefrontal cortex.

Visuospatial processing and response were assessed via the visual reproduction I and II subtests of the WMS-III INDIA and one independent test, the Shepard mental rotation task. Treatment with ashwagandha was not associated with statistically significant enhancement of these parameters compared with placebo after eight weeks ($p > 0.05$). Visuospatial processing and response are controlled by the posterior parietal cortex and middle/inferior frontal gyrus (de Graaf et al., 2010). It may be observed that ashwagandha exerts minimal effects in these regions of the brain.

Executive function was assessed using the Wisconsin card sort test and the Eriksen Flanker task. Treatment with ashwagandha was associated with enhanced executive function parameters measured by these instruments as indicated by a significant increase in these scores compared with placebo after eight weeks ($p < 0.05$).

Treatment with ashwagandha was also associated with significant enhancement of attention and information processing speed compared with placebo, as indicated by the scores on the trail-making test part A and the Mackworth Clock test at baseline and after eight weeks ($p < 0.05$).

The present study has several strengths. These include its prospective design, the fact that subjects were randomized to receive treatment or placebo control, and that the treatment was double-blinded. Limitations include small sample size due to the study's status as a pilot study. A larger clinical trial with longer follow-up is needed to confirm the promising results of this study.

Conclusions

Ashwagandha (*Withania somnifera* (L.) *Dunal*) may be useful in enhancing immediate and general memory, executive function, attention, and information processing speed in people with MCI, with few adverse effects.

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Declaration of interest

The authors declare that there are no conflicts of interest. The authors alone are responsible for the content and writing of this article.

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