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Research Article

Alpinia galanga Extract Increases Alertness, Focus, and Energy While Lowering Fatigue and Daytime Sleepiness with Four Weeks Supplementation: A Randomized, Double-Blind, Placebo-Controlled, Cross-Over Study in Human Subjects

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Abstract

Introduction: Nootropics are substances that enhance cognitive performance. Natural nootropics are preferred since they have fewer side effects and are effective and safe for long-term use. *Alpinia galanga* is an herbal ingredient that has been studied extensively as a supplement for improving mental alertness, focus, and energy. **Methods:** The present study is a randomized, double-blind, placebo-controlled, cross-over, clinical interventional study to evaluate efficacy and safety of *A. galanga* extract (AGE), commercially known as EnXtra®. A total of 62 subjects were randomized to receive either placebo or AGE as per the randomization schedule. Subjects were instructed to consume one capsule every day 30 minutes after lunch for 28 days in period I and II. A washout period of 10 days was followed between the two periods. The primary endpoint of the study was mental alertness assessed by the Symbol Digit Coding (SDC) test, Shifting Attention (SAT) test, Stroop Test (ST), and Alertness Rating Scale (ARS) of CNS Vital Signs; and secondary endpoints evaluated daytime sleepiness and fatigue, energy, and vigor, respectively, through the Epworth Sleepiness Scale (ESS), and the Visual Analogue Scale to Evaluate Fatigue Severity (VAS-F) at baseline and day 28 of supplementation. Safety was evaluated by monitoring adverse events, hematology and clinical chemistry tests. The results were

compared between AGE and Placebo groups. **Results:** Sixty-one subjects from the placebo group and 60 subjects from the AGE group completed the study. Compared to baseline at day 28, the AGE group showed significant improvements ($p<0.05$) in correct responses based on SDC, SAT, and mean correct responses from SDC and SAT. Further, there was a significant reduction in errors based on SAT, and mean errors from SDC, SAT & ST; simple reaction time based on ST; and correct reaction time based on SAT. Additionally, AGE group showed significant increase in ARS scores and decrease in ESS scores; significant increase in VAS – Energy, and Vigor sub-scale scores and decrease in VAS – Fatigue score. No adverse events were observed related to the test products. **Conclusion:** Twenty-eight days of AGE supplementation demonstrated significant improvements in alertness, reaction time, accuracy, fatigue, energy and vigor as compared to placebo. No safety concerns were observed throughout the study.

Trial Registration: Clinical Trials Registry of India CTRI/2022/05/042770

Introduction

The quest for improved productivity in a technology-driven world today has increased the demand for nootropics that promise better focus, memory, clearer thinking, and increased productivity [1]. Starting the day with a drink of coffee is a common practice worldwide. Drinking coffee is known to increase alertness, improve mood, enhance vigilance, faster reaction times, and increase attention [2]. However, excessive coffee consumption may lead to jitters, anxiety, and sleep deprivation, whereas, avoidance may trigger withdrawal symptoms like headaches, drowsiness, decreased alertness, and fatigue. Further, individuals may have varying degrees of sensitivity to coffee, with possible caffeine dependence or addiction. Excessive coffee consumption can also cause tachycardia, palpitations, and elevated blood pressure [2-5]. Hence, there has been a constant search for safer and more effective natural psychostimulants as an alternative to coffee.

The rhizome of Alpinia galanga is a rich source of flavonoids, terpenoids, and essential oils and is known to induce psychostimulant activity. A. galanga is an aromatic herb widely used as a spice for flavoring foods in Asian countries and well known in traditional medicine for treating numerous health conditions [6-8]. Preclinical studies have demonstrated that *A. galanga* is associated with anti-amnesic and neuroprotective activity mediated through free radical scavenging activity, cholinesterase inhibition, and inhibition of pro-inflammatory cytokines [9,10]. Further aqueous extracts of A. galanga have been explored as a safer psychostimulant and found to be more efficient than caffeine in maintaining mental alertness without inducing the caffeine-crash-like symptoms in healthy young volunteers. Further, A. galanga extract (AGE) was found to be more effective than other plant extracts such as Cymbopogon flexuosus (lemongrass) and Glycyrrhiza glabra (licorice), with a significant and stable increase in alertness scores from baseline until 5 hours in healthy subjects [11]. Additionally, AGE by itself or in combination with 200 mg caffeine was able to improve mental alertness with sustained attention at 3 hours post dose [11-16]. A molecular docking study indicated that the neurocognitive-enhancing property of AGE is likely mediated by the interaction of bioactive components of the extract with various targets involved in dopamine and acetylcholinesterase pathways [12].

Further, we validated previous results that demonstrate the same-day nootropic benefits of AGE on improved alertness, accuracy, and attention as early as 30 minutes of supplementation and lasting up to five hours with a significant increase in subjective feelings of energy and decreased fatigue levels [17]. Here we report the extension of the same-day effect study on mental alertness and fatigue as above after continuation of supplementation with AGE for 28 days in the same subjects after re-randomization.

Methods

Study Design and Procedures

The present study was a randomized, double-blind, placebo-controlled, cross-over, clinical study on healthy males and females, aged between 18 and 55 years. The study was initiated after obtaining written approval from BGS Global Institute of Medical Sciences Institutional Ethics Committee, Bengaluru, India. The study was conducted as per the requirements of the Indian Council of Medical Research (ICMR) ethical guidelines, International Council for Harmonization (ICH) Guidance on Good Clinical Practice (E6R2) and the Declaration of Helsinki. The study was registered with the Clinical Trials Registry of India (CTRI/2022/05/042770).

A voluntary informed consent process was completed for each subject before enrolling them in the study. Visit 1 was the screening period (day -7 to day -1) where demographic details such as date of birth, sex, ethnicity, and race were obtained. A medical history and medication history were obtained. The eligible subjects were randomized at their next visit on day 1 to receive placebo or AGE as per the randomization schedule. After a washout period of 5 days, at visit 3 (day 7), subjects were crossed-over to receive placebo or AGE as per the randomization schedule. Visits 1 to 3 of the study were considered part I, i.e., the acute phase.

After the completion of the acute phase and a washout period of at least 5 days, part II (4 weeks of supplementation phase) of the study was conducted from visit 4, where the eligible study subjects were re-randomized to receive placebo or AGE for 28 days, followed by a follow-up visit (visit 5) on day 40. After a washout period of 10 days, at Visit 6, subjects were crossed-over to receive placebo or AGE for another 28 days as per the randomization schedule.

Baseline assessments were performed on Visit 4 (Day 13) and Visit 6 (Day 51), and Day 28 assessments were performed on Visit 5 (Day 40) and Visit 7 (End of Study Day 78) for the primary efficacy endpoints: Central Nervous System Vital Signs (CNS VS) - Symbol Digit Coding (SDC), Shifting Attention (SAT), Stroop Test (ST), and Alertness Rating Scale (ARS), and secondary efficacy endpoints: Epworth Sleepiness Scale (ESS) and Visual Analogue Scale to Evaluate Fatigue Severity (VAS-F) – Fatigue, Energy, and Vigor sub-scales.

The study product contained 300 mg of AGE (commercially known as EnXtra®) and 90 mg of microcrystalline cellulose, and the placebo contained 390 mg of microcrystalline cellulose in the form of scarlet-red opaque hard gelatin capsules (manufactured by OmniActive Health Technologies Ltd., Mumbai, India). Subjects were asked to consume one capsule every day for 30 minutes post-lunch for 28 days, with a cross-over of treatment at Visit 6.

Study population and eligibility criteria

Subjects who met all the following inclusion criteria were included in the study: healthy male or female adults, aged 18 to 55 years (both limits inclusive), had a BMI of 18.5 kg/m2 to 29.9 kg/m² (both limits inclusive), had a Fatigue Severity Scale score >4, had post-lunch sleepiness as indicated by ESS ≥11 and ≤17, and history of consuming <3 cups of tea/coffee per day, agreed to sleep for 8 ± 1 hours the night before the visit day, agreed to maintain their usual dietary habits and level of exercise i.e. maintain their usual life-style throughout the trial period, agreed to refrain from taking any medications or preparations to improve fatigue (herbal, dietary supplements, homeopathic preparations, etc.) during the study period, agreed to refrain from consuming alcohol 24 hours prior to the visit days, agreed to refrain from consuming caffeine and caffeine-containing products 12 hours prior to visit days, agreed to refrain from vigorous physical activity 12 hours prior to visit days, and agreed to stay weight stable during the study period.

Subjects who met any of the following criteria were excluded from the study: had hypersensitivity or a history of allergy to the study product, with moderate to severe fatigue or having chronic fatigue syndrome, with a malignant disease or any concomitant end-state organ disease and/or laboratory abnormalities considered by investigators to be risky or that could interfere with data collection, suffering from a metabolic disorder (uncontrolled diabetes, uncontrolled thyroidal condition) and/or from severe chronic disease (cancer, renal failure, HIV, immunodeficiency, hepatic or biliary disorders, arthritis, uncontrolled cardiac disease) or from a disease found to be inconsistent with the conduct of the study by the investigator, with a psychiatric diagnosis other than anxiety or depression, with sleep disturbances and/or were taking sleep aid medication, with uncontrolled hypertension (systolic blood pressure >160 mm Hg or diastolic blood pressure >100 mm Hg) at screening, who were on anxiolytics, anti-depressants, antipsychotics, anticonvulsants, antihypertensive, centrally acting corticosteroids, opioid pain relievers, hypnotics, and/or prescribed sleep medications, with a history of drug and /or alcohol abuse at the time of enrolment, who were pregnant, nursing, or planning a pregnancy within the study participation period, with positive Urine Pregnancy Test at Screening/Randomization Visit, who had been treated with any investigational drug or investigational device within a period of 3 months prior to study entry, and any additional condition(s) that in the Investigators opinion would warrant exclusion from the study or prevent the subject from completing the study.

Determination of Sample Size

The sample size was calculated based on the outcomes of symbol-digit coding measures from the published data to detect the difference between the two treatments. Assuming a common SD of 4.50 at the end of treatment, a total of 52 subjects were sufficient to detect a difference of 3.00 in the mean difference between the two treatments with a power of 90% and a 0.05 two-sided level of significance. A total of 62 subjects were enrolled in this cross-over study, assuming a 15% dropout rate.

General Statistical Considerations

For continuous endpoints, results were summarized using descriptive statistics: number of non-missing subjects (n), mean, standard error, minimum, median, and maximum. For evaluations involving the primary and secondary efficacy endpoints, change values and the mean change from baseline to the end of the study were computed. A paired t-test was applied to assess within-group analysis. An independent t-test was applied to assess between group analysis for the actual change and percentage change. A value of $p < 0.05$ was considered statistically significant.

Categorical variables were summarized using counts and percentages. The count [n] indicates the actual number of subjects with a value of a variable or event, which should always be less than or equal to the total number of subjects with a non-missing value of the variable or event in the respective study group [N]. The comparisons between placebo and AGE groups with the supplement capsule were evaluated using the Pearson's Chi-

Square test or Fisher's exact test, as appropriate. The analysis was conducted on the safety population only.

For inferential tests, a p-value < 0.05 and 95% confidence intervals were considered for statistical significance, and a two-tailed hypothesis was tested. All statistical analyses were conducted with R statistical software version 4.2.1 and were two-sided at a significant level of p < 0.05. Missing data were not included in the analyses. All subjects recruited into the study were accounted for, including those who did not complete the study, along with the reasons for withdrawal.

Efficacy Analysis

Primary efficacy endpoints included SDC, SAT, ST test, and ARS of CNS VS. The secondary efficacy endpoints were based on ES scores and VAS-F - fatigue, energy, and vigor subscale scores. The efficacy analyses were performed on all study subjects' data. Assessments were done at baseline, i.e., visits 4 and 6, and after 28 days of treatment, i.e., visits 5 and 7.

CNS VS is a computerized neuropsychological test to evaluate the neurocognitive status of subjects and covers a range of mental processes from simple motor performance, attention, memory, to executive functions. Mental alertness was assessed by SDC test, SAT test, ST and ARS of CNS Vital Signs. The results, from baseline and day 28, were compared between AGE and placebo group.

The secondary objectives of the study were to evaluate the effect of AGE compared to placebo on fatigue, energy and vigor as assessed by VAS-F and daytime sleepiness by ESS. Assessments were done at baseline i.e., visits 4 and 6, and after 28 days of supplementation i.e., visits 5 and 7.

For evaluations involving the primary and secondary efficacy endpoints, a paired t-test was applied to assess within group analysis (placebo and AGE). An independent t-test was applied to assess between-group analysis (AGE versus placebo) for change in actual values and mean change from baseline at the end of the study. A non-parametric test was used if the data was not normally distributed. A value of p<0.05 was considered statistically significant.

Safety analysis

The safety population includes all randomized subjects who received investigational products. All safety assessments, including adverse events, clinical laboratory test results, vital signs (blood pressure, heart rate, and body temperature), and physical examinations, were listed by subject and tabulated when applicable.

The continuous-type safety endpoints were summarized by treatment using descriptive statistics (n [number of subjects], mean, and percentage) on the safety population. The categorical-type safety endpoints were summarized by treatment using frequency and percentage of the safety population. The difference between the placebo and AGE groups was evaluated using the Pearson's Chi-Square test or Fisher's exact test, as appropriate. The analysis was conducted on the safety population only.

Demographic and baseline characteristics such as age, height (cm), weight (kg), and body mass index (BMI) (kg/m2) were summarized and tabulated by randomized treatment group and overall. The analysis was based on the randomized population.

Descriptive statistics (n, mean, standard deviation, median, minimum, and maximum) were presented for age (derived relative to informed consent date), height, weight, and BMI. Frequency counts and percentages were presented for gender. All individual subject demographic and baseline characteristic data were listed.

Results

A total of 68 subjects were screened, of whom 6 subjects were screen failures (Figure 1). A total of 62 subjects were re-randomized into the 4-week supplementation phase of the study on day 13 after the completion of the acute phase. Of the 62 re-randomized subjects, 61 subjects completed the placebo group as one subject withdrew consent due to personal reasons, and 60 subjects completed the AGE group as 2 subjects withdrew consent due to personal reasons and were included for the efficacy assessments. All 62 subjects were included in the safety assessment.
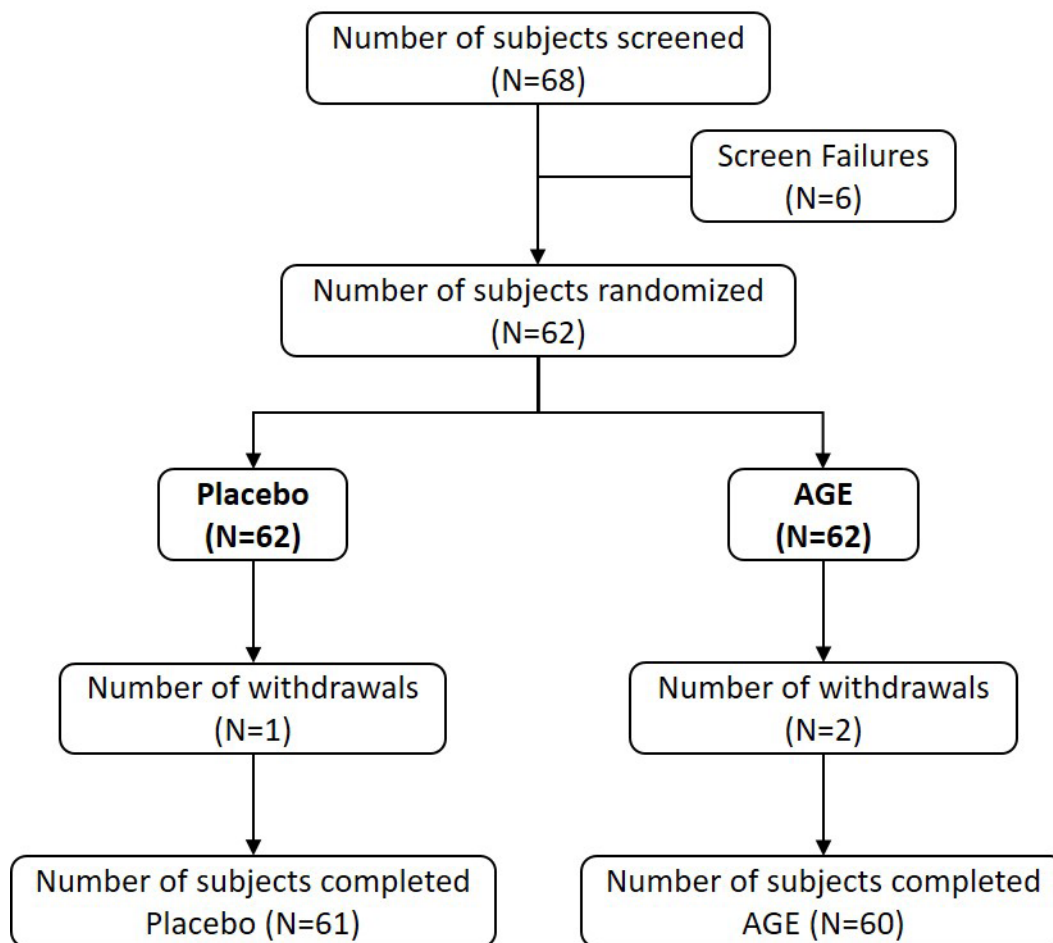
Figure 1: CONSORT diagram - – Alpinia galanga Extract Four Week Supplementation Phase Study.

Subjects' demographics and baseline data are presented in Table 1. The mean (±SD) age of subjects in placebo group was 35.21 ± 6.42 years, and BMI was 23.89 ± 2.06 Kg/m2 and in AGE group was 35.35 ± 6.51 years, and BMI was 23.78 ± 1.97 Kg/m2. Study population consisted of 60% males and 40% females.

Table 1 Subject demographics

Parameters	Statistics	Placebo (N=61)	AGE (N=60)
Gender: Male	n (%)	36 (59%)	36 (60%)
Gender: Female	n (%)	25 (41%)	24 (40%)

Parameters	Baseline (N=62)	End of Study	
		Placebo (N=61)	AGE (N=60)
Age (years)			
Mean	35.35	35.21	35.35
SD	6.43	6.42	6.51
SE	0.82	0.82	0.84
Minimum	21	21.00	21.00
Median	35.50	35.00	36.00
Maximum	46	46.00	46.00
Weight (kg)			
Mean	66.23	65.59	65.25
SD	10.75	10.09	9.88
SE	1.37	1.29	1.28
Minimum	45.00	44.70	44.70
Median	68.10	66.40	66.15
Maximum	89.70	87.20	87.20
Height (cm)			
Mean	165.12	165.31	165.25
SD	9.89	9.95	9.99
SE	1.26	1.27	1.29
Minimum	145	145.00	145.00
Median	165	165.00	165.00
Maximum	183	183.00	183.00
BMI (Kg/m²)			
Mean	24.16	23.89	23.78
SD	2.25	2.06	1.97
SE	0.29	0.26	0.25
Minimum	19.10	19.90	19.90
Median	24.30	24.00	23.90
Maximum	28.60	29.20	29.20

% - Percentage; AGE - *Alpinia galanga* extract; N - Number of subjects; SD - Standard deviation; SE - Standard Error.

Efficacy

Primary efficacy endpoints

i. Symbol Digit Coding (SDC) Test - Correct Responses and Errors

A statistically significant ($p < 0.05$) increase in correct responses was recorded in the AGE group compared to the placebo group at day 28 (8.25 ± 2.51 in AGE group and -8.10 ± 2.20 in placebo group; Fig. 2a).

However, no significant difference was observed between AGE and placebo groups for errors at day 28 (-0.07 ± 0.25 in AGE group and 0.49 ± 0.27 in placebo group; Fig. 2b).

ii. Shifting Attention Test (SAT) - Correct Responses, Errors and Correct Reaction Time

The AGE group showed a statistically significant increase in correct responses (11.52 ± 1.49 in AGE group and -2.54 ± 1.71 in placebo group, as shown in Fig. 2c), as well as a statistically significant decrease in errors (-5.47 ± 0.66 in AGE group and 0.03 ± 0.81 in placebo group, as shown in Fig. 2d) and correct reaction time (-69.88 ± 29.07 in AGE group and 74.54 ± 27.65 in placebo group, as shown in Fig. 2e), on day 28 as compared to the placebo group.

iii. Stroop Test (ST) - Simple Reaction Time, Complex Reaction Time: Correct, Stroop Reaction Time: Correct and Stroop Commission Errors

The AGE group showed a significant ($p < 0.05$) decrease in simple reaction time as compared to the placebo group on day 28 (-108.03 ± 30.26 in AGE group and 35.44 ± 36.96 in placebo group; Fig. 2f).

A significant ($p < 0.05$) decrease in complex reaction time: correct was recorded between the two groups compared to baseline at day 28 (-47.13 ± 37.39 in AGE group and 67.90 ± 30.89 in placebo group; Fig. 2g), however, there was no statistical significance in AGE group as compared to baseline ($p = 0.2124$).

No significant difference was recorded between the two groups for Stroop reaction time: correct at day 28 (-26.07 ± 27.49 in AGE group and 39.07 ± 26.32 in placebo group; Fig. 2h).

A decreasing trend ($p = 0.0736$) in the Stroop commission errors was recorded in the AGE group as compared to the placebo group at day 28 (-0.77 ± 0.23 in AGE group and 0.14 ± 0.45 in placebo group, Fig. 2i).

iv. Mean correct responses scoring from Symbol Digit Coding (SDC) and Shifting Attention Test (SAT)

A significant ($p < 0.05$) increase in the mean correct responses scoring from SDC and SAT was recorded in the AGE group compared to the placebo group at day 28 (9.88 ± 1.73 in AGE group and -5.32 ± 1.47 in placebo group; Fig. 3a).

v. Mean errors scoring from Symbol Digit Coding (SDC)-Errors, Shifting Attention Test (SAT)- Errors and Stroop Test (ST)- Stroop Commission Errors

A significant ($p < 0.05$) decrease in the mean errors scoring (from SDC - Errors, SAT - Errors and ST - Stroop Commission Errors) was recorded in the AGE group compared to the placebo group at day 28 (-2.09 ± 0.27 in AGE group and 0.22 ± 0.35 in placebo group; Fig. 3b).

vi. Mean correct reaction time from Shifting Attention Test (SAT)- Correct Reaction Time and Stroop Test (ST)- Complex Reaction Time Correct, Stroop Reaction Time Correct

A significant ($p < 0.05$) decrease in the mean correct reaction time scoring (from SAT - Correct Reaction Time and ST-Complex Reaction Time Correct, Stroop Reaction Time Correct) was recorded in the AGE group compared to the placebo group at day 28 (-47.69 ± 22.82 in AGE group and 60.50 ± 19.88 in placebo group; Fig. 3c).

vii. CNS Vital Signs: Alertness Rating Scale (ARS) scoring

A significant ($p < 0.05$) increase in the ARS scoring was recorded in the AGE group compared to the placebo group at day 28 (1.07 ± 0.15 in AGE group and -0.23 ± 0.12 in placebo group; Fig. 3d).

Secondary efficacy endpoints

i. Epworth Sleepiness Scale

A significant ($p < 0.05$) decrease in ESS scoring was recorded in the AGE group compared to the placebo group at day 28 (-3.32 ± 0.52 in AGE group and -1.03 ± 0.43 in placebo group; Fig. 3e).

ii. Visual Analogue Scale (VAS) to evaluate Fatigue Severity

Fatigue Score

The AGE group exhibited a significant ($p < 0.05$) decrease in the VAS - Fatigue Score as compared to the placebo group at day 28 (-5.60 ± 0.42 in AGE group and -0.73 ± 0.46 in placebo group; Fig. 3f).

Energy Score

The AGE group showed a significant ($p < 0.05$) increase in the VAS - Energy score as compared to the placebo group at day 28 (4.77 ± 0.56 in AGE group and 2.13 ± 0.56 in placebo group; Fig. 3g).

Vigor Sub-scale Score

A significant ($p < 0.05$) increase in the VAS - Vigor Score was recorded in the AGE group as compared to the placebo group at day 28 (3.17±0.84 in AGE group and 0.82±0.79 in placebo group; Fig. 3h).
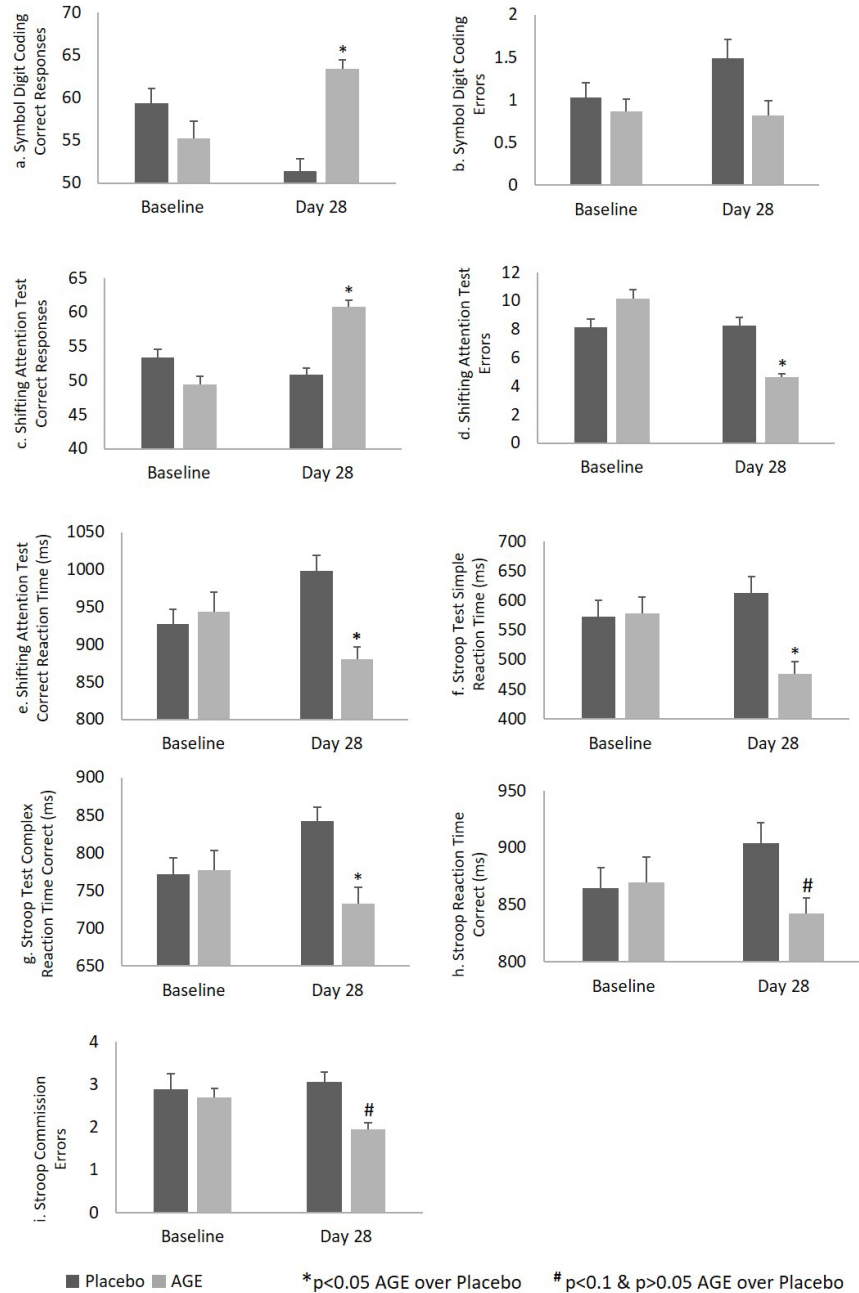
Figure 2: Summary of results Symbol Digit Coding (SDC) test – Correct Response (a) and Errors (b); Shifting Attention (SAT) test – Correct Responses (c), Errors (d), Correct Response Time (e); Stroop Test (ST) – Simple Reaction Time (f), Complex Reaction Time Correct (g) and Stroop Reaction Time Correct (h) and Stroop Commission Errors (i) at baseline and day 28 of treatment. AGE – *Alpinia galanga* Extract; * $p < 0.05$ AGE over Placebo.
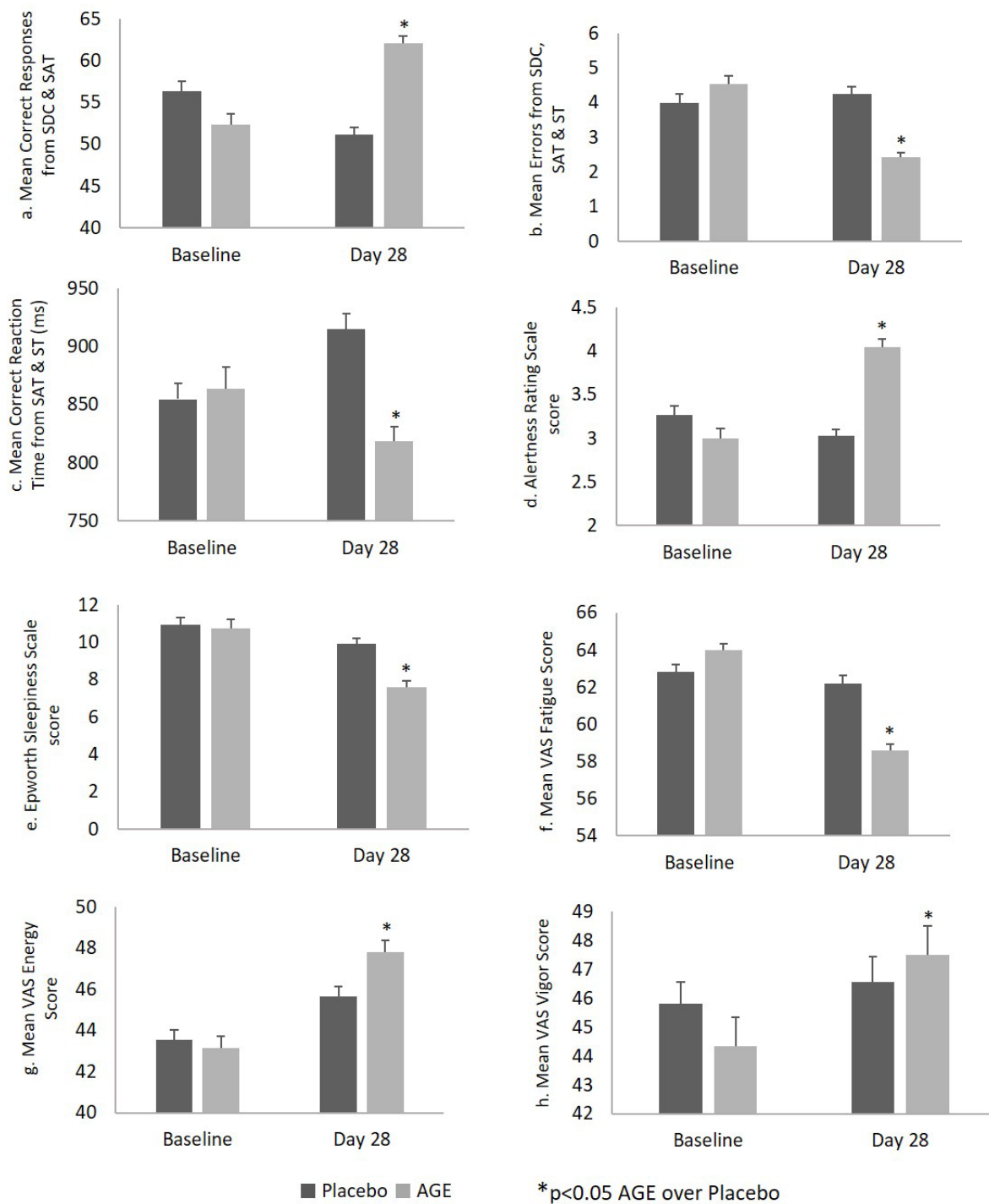
Figure 3: Summary of mean Correct Responses from SDC and SAT (a); Mean Errors from SDC, SAT and ST (b); Mean Correct Reaction Time from SAT and ST (c); Alertness Rating Scale (ARS) (d); Epworth Sleepiness Scale (ESS) (e); and Visual Analogue Scale (VAS) to Evaluate Fatigue Severity – Fatigue (f), Energy (g) and Vigor (h) at baseline and day 28 of treatment. AGE – *Alpinia galanga* Extract; * p<0.05 AGE over Placebo. # p<0.1 and p>0.05 AGE over Placebo.

Safety Results

No clinically significant changes were observed in the hematology parameters and liver function parameters (Aspartate aminotransferase (AST), Alanine transaminase (ALT), Bilirubin), kidney function parameters (Serum creatinine, blood urea nitrogen) and vital sign measures like blood pressure, heart rate, and body temperature throughout the study. During the course of the study, a total of 12 adverse events (AEs) were reported by 6 subjects. Two subjects (3.23%) during AGE administration experienced 5 AEs and 4 subjects (6.45%) during placebo administration experienced 7 AEs. Subjects in AGE group reported AEs – headache (2), fever (1), viral fever (1), cold (1) and subjects in the placebo group reported AEs – allergic rhinitis (1), headache (2), fever (1), cold (3).

All the AEs reported by the subjects were mild in severity and the causality of the AEs was diagnosed by the investigator as not related to the investigational products. None of the subject reported any SAE or was withdrawn from the study due to an AE or SAE.

Two subjects from the AGE group and 4 subjects from placebo group used at least one concomitant medication during the course of the study. Most commonly used medication included paracetamol.

Discussion

Previous studies conducted in human subjects demonstrated that AGE promotes alertness after oral consumption starting from 30 minutes lasting up to 5 hours, unlike caffeine, which rapidly promotes mental alertness within the first hour followed by a steep decline thereafter [15]. We further validated the same-day nootropic benefits of AGE using a larger subject population and demonstrated that AGE supplementation significantly improved mental energy, including mental alertness, attention, reaction time, correct responses, and reduction in errors, as early as 30 minutes and sustained until five hours post-dose. Additionally, AGE demonstrated a significant increase in subjective feelings of energy and decreased fatigue levels [17]. In the current study, we report the extension of the above same-day effect study and demonstrate the nootropic effect of AGE over long-term use. We re-randomized the subjects who participated in the acute phase of the study and continued the supplementation of AGE at 300 mg per day or placebo for 28 days, followed by a washout period of 10 days and cross-over as per the randomization schedule. We observed that supplementation with AGE for 28 days resulted in significant improvements in cognitive functions such as improved alertness and accuracy, a decrease in daytime sleepiness and fatigue, and increased energy and vigor levels. We also observed that, throughout the study period, AGE supplementation was well

tolerated with no negative side effects and thus safe to consume for a longer duration.

CNS Vital Signs tests are used for assessment of neurocognitive functions such as processing speed (SDC), executive function (SAT), reaction time (ST) and alertness (ARS) [17-19]. We observed statistically significant ($p<0.05$) outcomes for some of the primary efficacy points investigated, such as increased correct responses (SDC Test), decreased errors (SAT), shorter simple and complex reaction time (ST), and increased alertness rating score (CNS vital signs) for the AGE group as compared to the placebo on day 28. This is in agreement with our previously reported acute study outcome, where we observed improved neurocognitive functions as early as 30 minutes after intake and lasting for up to 5 hours [17]. Similarly, we observed a significant improvement in secondary efficacy endpoints such as decreased fatigue, increased energy and vigor as assessed by VAS-F, and reduced daytime sleepiness as measured by ESS in response to AGE supplementation on day 28. The nootropics act as calcium channel blockers, acetylcholinesterase inhibitors, serotonergic, dopaminergic, and glutamic acid receptor antagonists, and antioxidants. Further, nootropics provide neuroprotection by reducing Aβ accumulation, synaptic dysfunction, inflammation, and oxidative stress [20,21], as well as improve neuroplasticity leading to enhanced cognitive functions [1]. *A. galanga* extract is rich in phytochemical components such as 1′S-1′-acetoxychavicol acetate and galangin with established neuroprotective function under experimental conditions [22-25]. AGE increases dopamine levels either by blocking dopamine uptake in the neuronal synapse or by inhibiting the enzyme Catechol-O-Methyltransferase, which is responsible for the degradation of dopamine in synaptic space (unpublished data). Thus, we believe that AGE may possibly increase dopamine levels in synapses and help to improve visuospatial performance and mental alertness. The AGE used in this study is an aqueous extract of A. galanga with a phytochemical content of 30% glycosides, 3% polyphenols, 3% flavonoids, and 1% tannins. Future studies should focus on detailed molecular analysis of the extract for a better understanding of the molecular mechanism of AGE leading to nootropic benefits including alertness, focus, and energy.

Unlike synthetic nootropics that may cause undesirable side effects, [26] natural nootropics are relatively safer when consumed even at higher doses and over a longer duration [27-29]. Similar to our previous acute phase study [17], four weeks of supplementation with AGE at a dose of 300 mg per day showed no adverse effects throughout the study duration.

The present study is constrained by the absence of control over several factors that contribute to fatigue, including but not limited to social life, work hours, and work-related stress.

Nevertheless, we implemented a standardization process for the subjects included in the study, which involved establishing specific criteria for fatigue levels. Another limitation is that the study was designed to measure nootropic benefits for only one hour on day 28, so the benefit of daily supplementation with AGE for 5 hours has yet to be determined.

Conclusion

Our study results conclude that AGE demonstrated significant improvements in cognitive function over placebo, as measured through various validated assessments. AGE could be beneficial for both short- and long-term consumption in improving mental functions, which makes it a potential alternative to other nootropics. Also, AGE was well tolerated and safe to consume for a longer duration.

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Author Contributions: MME, MK, JT contributed to the study conception and design. Study conduct, subject recruitment and data collection were performed at respective study centres by MME & MK. Study was monitored in blinded fashion by LJ. Statistical analysis and study report were prepared by JT. Data interpretation was done by MME, MK and JT. The first draft of the manuscript was written by MME, MK and JT. All authors provided their inputs, read and approved the final manuscript.

Disclosures: The authors report there are no conflicting interests to declare.

Compliance with Ethics Guidelines: A written approval was obtained from the Ethics Committees of BGS Global Institute of Medical Sciences Institutional Ethics Committee and Divakars Speciality Hospital Ethics Committee, Bengaluru, India, before the commencement of the study. The study was registered with the Clinical Trials Registry of India (CTRI/2022/05/042770). The study was conducted as per the regulatory requirements of the Indian Council of Medical Research, ethical guidelines, the International Council for Harmonization Guidance on Good Clinical Practice (E6R2), and the Declaration of Helsinki. A voluntary informed consent was obtained in writing from every participant before enrolling in the study.

Data Availability: The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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