Research Article **Open-Access**

Clinical evaluation of a novel gallotannin-enriched Galla Rhois extract (GRE) on vital cognitive functions in healthy volunteers: A randomized, double-blind, placebo-controlled study

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Submission Date: August 25th, 2023; **Acceptance Date:** September 6th, 2023; **Publication Date:** October 16th, 2023

Please cite this article as: Heuer M., Baker C., Sedlak M., Woo K. J., Kee K. H., Bagchi D. Clinical evaluation of a novel gallotannin-enriched Galla Rhois extract (GRE) on vital cognitive functions in healthy volunteers: A randomized, double-blind, placebo-controlled study. *Functional Foods in Health and Disease* 2023; 13(10):487-504. DOI: https://www.doi.org/10.31989/ffhd.v13i9.1213

ABSTRACT

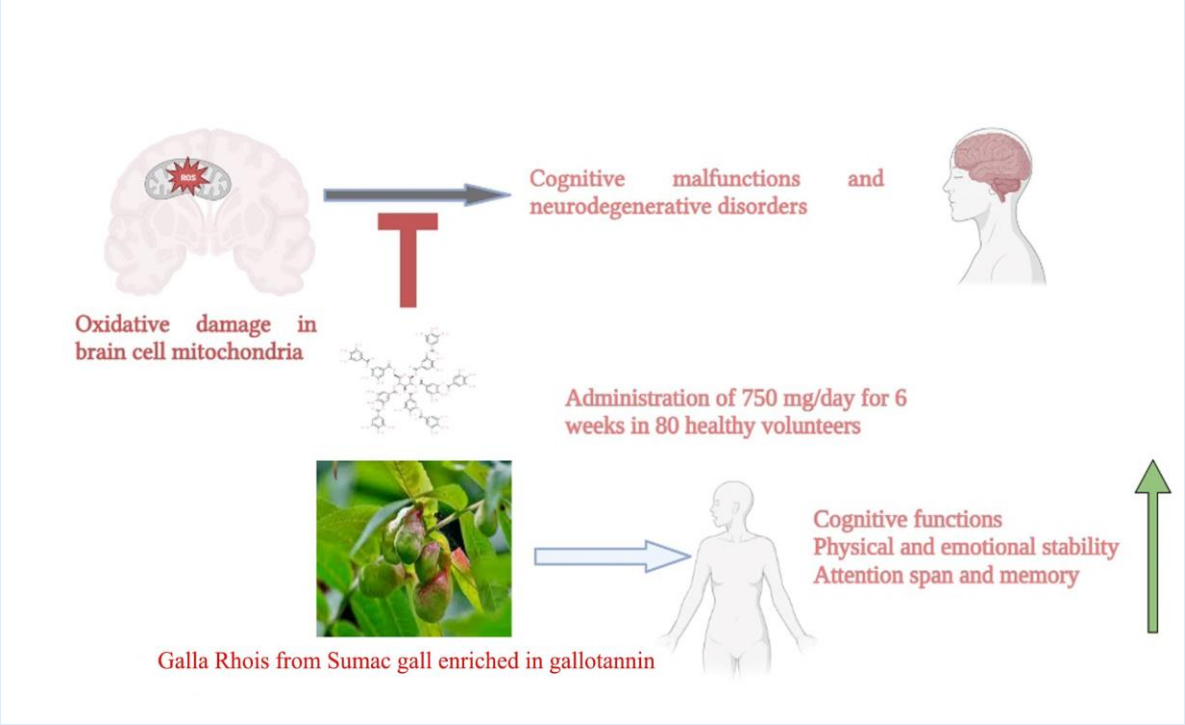
Background: Oxidative damage has emerged as one of the significant factors in the onset of neurodegenerative disorders, primarily owing to a) higher usage of oxygen by brain cells and b) higher amounts of lipids in the neuronal membranes. Together, these have resulted in higher susceptibility of brain cells to ROS-induced lipid peroxidation, damage to other cellular macromolecules, including nucleic acids and proteins, and death of neurons by apoptosis, ferroptosis, and necrosis. The design of therapeutics to prevent oxidative damage-induced brain stroke or other irreversible injuries has remained a challenge owing to the continuous production of free radicals at the site of damage. In this respect, plant-derived tannins and tannic acid derivatives have exhibited promising results in scavenging free radicals and inhibiting lipid peroxidation.

Objective: This investigation reports the effect of Ghala Rois extract (GRE), a methyl gallate enriched gall formed on the nutgall sumac tree, Rhus javanica L. (Anacardiaceae), on enhancing the cognitive function and overall well-being through a double-blind, placebo-controlled, randomized, parallel-group design study on 80 healthy adult volunteers over a period of six consecutive weeks.

Results: Administration of GRE in the dosage of 750 mg/day p.o. (with an equal amount of corn starch as a placebo) after breakfast resulted in significant improvement in cognitive functions as assessed by HappyNeuroPro, a digital therapy tool, and the cognitive tools under Mini-Mental State Examination (MMSE), Montreal Cognitive Assessment (MOCA) and Adult ADHD Self-Reported Scale (ASRSv1.1). Concomitantly, the GRE-treated subjects also scored significantly higher ($p = 0.033$) in the roles of Physical and Emotional Problems, Energy/Fatigue, Emotional well-being, and pain as evaluated by linearly independent pairwise comparisons among the estimated marginal means and computed using alpha = 0.05. In the Global Evaluation Questionnaire, over 77% of the treated subjects believed that the product increased their attention span, and helped their memory by 70%, and they also indicated their willingness to buy the commercially available GRE. Evaluation of the complete metabolic profile in the GRE-treated subjects significantly lowered the blood urea nitrogen (BUN) (11.98 mg/dL) as compared to placebo (13.27 mg/dL) ($p = 0.011$) as well as SGPT/ALT from 28.09 U/L (placebo) to 18.17 U/L (GRE-treated) ($p<0.001$). However, the vital parameters, including heart rate and systolic/diastolic pressure, were not affected by the consumption of GRE extract.

Conclusion: The study demonstrated that GRE could be used as an effective new phytotherapeutic for reversing oxidative damage-induced neuronal degeneration and improving cognitive health in these study individuals.

Keywords: Galla Rhois extract (GRE); Rhus javanica L. (family Anacardiaceae); Cognitive functions; ADHD; Executive function; Verbal memory; Safety, Reactive Oxygen Species (ROS)



Footnote: Clinical investigation was conducted by Global Clinicals, Inc., Los Angeles, CA (*www.globalclinicals.com*)

INTRODUCTION

Oxidative stress has been identified as one of the most significant contributing factors to the development of neurodegenerative disorders [1]. Brain cells need a surplus amount of oxygen [2] as compared to other tissues and are the second highest reservoir of lipids in the body after adipose tissue [3]. The neuronal membranes are especially rich in Polyunsaturated Fatty Acids which act as potent centers to induce production of reactive oxygen species (ROS) leading to lipid peroxidation [4]. In addition to ROS, reactive nitrogen species (RNS) [1], and carbon- and sulfur-centered radicals [5] are also produced because of incessantly operating oxidative phosphorylation in mitochondria which cause substantial damage to cellular macromolecules including nucleic acids, proteins, and membrane phospholipids. Oxidative modifications in key cellular proteins like alpha-synuclein foster their amyloidogenic aggregation leading to the formation of plaques and neurofibrillary tangles enriched in beta-pleated sheets [6]. Even bigger implications come from impaired removal of damaged mitochondria by autophagy, a phenomenon termed mitophagy [7].

The persistence of damaged mitochondria increases the ROS load leading to activation of pro-apoptotic caspases and concomitant cell death by apoptosis. Apart from apoptosis, ferroptosis is another newly identified and increasingly significant iron-dependent mechanism of neuronal cell death that involves oxidation-induced gross changes in the phospholipid membrane mediated by lipid-ROS such as the lipid hydroperoxides [8]. Free iron in its ferrous state reacts with hydrogen peroxide, leading to the generation of highly reactive hydroxyl groups which in turn causes drastic changes in mitochondrial membrane permeability and concomitant cell death by acute energy depletion. Ferroptosis has been traced to be associated with many human ailments including tumorigenesis, cardio-vascular diseases (CVD), and ischemia-reperfusion injury (IRI) [9]. Chronic oxidative damage leading to progressive loss of neurons causes progressive loss of cognitive functions eventually leads to neurodegenerative disorders like Parkinson's disease (PD), Alzheimer's disease (AD), Huntington's Disease, and Amyotrophic Lateral Sclerosis (ALS) [10-12].

Most of the treatment strategies to arrest oxidative damage to brain cells include suppression, sequestration, and/or degradation of free radicals. However, lead compounds that had shown promising results with respect to one or more roles of the above in pre-clinical trials carried out in cell lines have failed to achieve desired effects in the clinical trials [13] plausibly due to limited blood-brain-barrier (BBB) permeability [10]. In addition, the extent and progression of the ROS-mediated reactions are often so fast that once initiated beyond the threshold value of cellular defense, there is practically no chance of arresting them with therapeutics. Phytochemicals are a powerful group of therapeutic molecules produced by plants as secondary metabolites which have been applied successfully for the treatment of a plethora of human ailments ranging from cancer to neurodegenerative disorders compounds [14].

The scope of functionality of phytochemicals in brain cells is manifold and versatile including modulation of neurotransmitter secretion and metabolism [15], controlling mitochondrial homeostasis [16] and acting as effective antioxidants to dampen free radical-induced oxidative stress [17]. However, probably the most promising part of the potential of phototherapeutics owe to their ability to regulate mitochondrial apoptotic machinery by inducing the anti-apoptotic Bcl-2 family signaling pathway and preventing amyloidogenic aggregation of proteins [17]. Over the last few decades, the major classes of phytochemical compounds with major neuroprotective potential have been polyphenols and other derived phenolics like tannins, the terpenes, &

alkaloids [18]. Galla rhois is a unique gall formed by the Chinese sumac aphid, *Schlechtendalia chinensis* (Bell), on the nutgall sumac tree, *Rhus javanica* L. (Anacardiaceae). It's extract (GRE), chemically, gallotannin polymers formed by esterification of gallic acid with a polyol carbohydrate have demonstrated anti-tumor [19], antioxidant [20], anti-inflammatory [21] and anti-viral activities [22].

Due to its strong antioxidant potential, Galla Rhois Extract (GRE) has also yielded encouraging results as a neuroprotectant in a rat model [23]. The prospect of Galla Rhois Extract (GRE) in preventing neurodegeneration is also propelled by the observation that gallotannins can effectively prevent aggregation of amyloid beta peptides implicated in AD [24]. However, not many studies have been conducted thus far on the potential of Galla Rhois Extract (GRE) in improving cognitive function in human trials. To the best of our knowledge, this is the first randomized double-blind placebo controlled report on the effect of consumption of Galla Rhois Extract (GRE) on improvement of cognitive abilities and motor functions in adult human models.

MATERIALS AND METHODS

A Novel Gallotannin-Enriched Galla Rhois Extract (GRE): This study determined the potential of a novel Galla Rhois extract (GRE, Manufacturer: Louis & Harry Biomed, Inc., Denver, CO) enriched in 1.3% gallic acid, which was based on a unique ethanolic extraction from aqueous phase a GMP certified manufacturing plant. The processing and handling and study were all conducted under proper GLP, GMP and GCP.

Subject Recruitment, Assignment and Ethical Approval: Healthy volunteers were recruited from the clinical database, online advertising, and local newspaper advertisements. Telephone prescreening was conducted prior to rescheduling the site visit and clinical examination. Subject recruitment procedures were performed using rigorous inclusion and exclusion criteria including medical examination as outlined in Table 1. All recruited subjects duly signed the informed consent after understanding the Bill of Rights. The clinical practices, experimental procedures, and study methodologies of this randomized placebo-controlled, double-blind study was performed in compliance and accordance with International Council for Harmonization (ICH) guidelines for Good Clinical Practices (GCP), including the archiving of essential documents as per International Ethical Standards guaranteed by the Declaration of Helsinki and its subsequent amendments. All under HIPAA compliance. Subject confidentiality was strictly enforced throughout the study. The Institutional Review Board (IRB) of the Ethical & Independent Review Services (Lee's Summit, MO, USA) approved this study protocol [Protocol #5547-6-0721; E&I Study Number 22057-01; E&I IRB #2:IRB00007807 Approval Date: Mar 29, 2022; Study Title: "Clinical Evaluation of a Galla Rhois Extract on Cognitive Function in Healthy Volunteers].

Subject Information and Informed Consent: Before the initiation of the study, the details of the IRB-approved study protocol along with the exclusion and inclusion criteria were clarified to the recruited study group subjects and asked to sign off the IRB-approved consent forms. All recruited and enrolled subjects read, understood, and gave their consents for the health questionnaire and the Consent Form by putting their signatures. Subject confidentiality was strictly enforced. Adverse event monitoring was strictly ascertained.

Study Compliance: The capsules corresponding to the placebo and treatment groups (GRE) were handed over to the respective subjects under each study group. All data were monitored and entered by the study

coordinator(s) and endorsed by the principal investigator (PI). Moreover, the study associate in charge of handing over the capsules carried out a double-check before signing off the investigational product (IP) accountability log.

Allocation Concealment: In order to ensure that there was no selection bias in the randomized control trial, allocations to study groups were concealed from the persons in charge of assessment of subjects taking the trial. In order to ensure concealment, opaque capsules were sequentially numbered in 84 capsule pack sealed white plastic bottles.

Study Discontinuation Clause: The criteria for the "stopping" of trial or "discontinuation criteria" was only in the case of serious adverse events (as defined in safety assessments clause).

Study Participants: A stringent IRB-approved inclusion and exclusion criteria (Table 1) was conducted and a total of 80 healthy male and female subjects (age: 20-55 years) were enrolled and randomly assigned into placebo and treatment (GRE) groups. As a double-blind study, the randomization codes were generated by SAS procedure PROC PLAN using block design. The recruited subjects were asked not to alter their daily physical activities. Tables 2A and 2B exhibit the demographic data and vital functions data of all enrolled subjects in the placebo and treatment (GRE) groups.

Table 1. Inclusion and exclusion criteria

A. **Inclusion criteria**	B. **Exclusion criteria**
Able to read, understand, and sign an Informed Consent.	Prior history of adverse reactions to any of the ingredients in the study preparations.
Cooperative, able to read, understand and answer questionnaires, and able to adhere to the study schedule.	Concomitant medication deemed by the clinical investigator to potentially interfere with or confound the study results. Examples of these classes of drugs are antidepressants, lithium, antipsychotics, anti-seizure medications, antihistamines and beta-adrenergic blockers. Any supplements for energy or mental enhancement.
Male and female volunteers (age: 20-55 years).	Any metabolic disorder including known electrolyte abnormalities that are not treated and stable.
In general, good health as demonstrated by medical history and affirmed by PI screening.	Contraindications such as severe constipation, pneumonia with coughing and other febrile diseases.
Willing to follow study instructions, complete all study visits, complete and submit any paperwork or questionnaires given.	Heart disease, arrhythmias, diabetes, thyroid disease, a history of hypertension, hepatorenal, autoimmune, or neurologic disease, or history of malignancies that are not treated and stable or history of malignancies other than treated basal cell carcinoma.
Willing to limit caffeine intake per Study Protocol.	Current or history of chronic alcohol and/or drug abuse.
Don't have severe cognitive function disorders (such as dementia, amnesia, delirium).	Participation in another clinical study within the past 30 days.

A. **Inclusion criteria**	B. **Exclusion criteria**
Willing to give blood samples via venipuncture.	Simultaneous participation in another clinical trial.
Willing to take study products (via oral delivery).	Any condition which, in the opinion of the investigator, makes the subject unsuitable for inclusion.
Valid contact information. If a change occurs, immediately contact staff. Agree to comply and follow study requirements.	Currently on any anticoagulant or antithrombotic medications (e.g., heparin, warfarin) All female subjects will undergo a urine pregnancy test before enrolling in the study unless they are post-menopausal or surgically sterilized. If positive, they will be excluded from the study. Also, women that are pregnant or planning on becoming pregnant will be excluded from the study.
Immediately report any SAE (side effects and/or adverse events) to study site staff or PI.	
Willing to follow COVID-19 safety precautions.	

All the study participants were instructed to consume either Galla Rhois Extract (GRE) or placebo capsules, without knowledge of which group. Treatment (GRE) group was instructed to consume 2 capsules of 375 mg (total 750 mg) each/day (standardized for >1.3% gallic acid), 30 minutes after the breakfast, while the placebo was asked to consume 2 capsules containing 375 mg of corn starch each, 30 minutes after the breakfast, over a period of six consecutive weeks. Both the placebo and GRE capsules were identical looking and same weight, respectively. Also, the placebo and GRE-treated subjects maintained daily diaries and get it regularly endorsed by the study coordinators. Adverse event monitoring was strictly enforced.

Demographics and Vital Functions: In this investigation, female: male ratio placebo group was 32.5: 67.5, while in the treatment (GRE) group the female: male ratio was 51.3 : 48.7. Both the groups were evenly distributed age-wise. Demographics and vital functions were compared with independent t-tests for means and Chi-square test for proportions. The following variables: (a) age, (b) body weight, (c) height, (d) BMI, and (e) gender are reported in Table 2A, while heart rate (HR), and systolic and diastolic blood pressure are exhibited in Table 2B. No significant differences were observed between the two groups for any of the variables.

Table 2A. Demographic data, heart rate and blood pressure of the study population

Data (Mean ± S.D.)	**Placebo** **N = 40**	**GRE** **N=40**	***p*-value**
Age (years)*	33.15 ± 10.15	33.65 ± 8.84	0.82
Body Weight (lbs)*	67.19 ± 4.44	65.79 ± 3.59	0.13
Height (in)*	178.50 ± 47.70	169.95 ± 32.32	0.35
BMI*	27.47 ± 5.19	27.51 ± 4.07	0.97

*Values are mean (standard deviation)

Table 2B. Vital functions: heart rate and blood pressure of the study population

Data (Mean ± S.D.)	Placebo N=40	GRE N=40	*p*-value
Heart rate (/min) *	75.87 (73.22 – 78.52)	76.73 (73.97 – 79.30)	0.69
Systolic bp (mmHg)*	122.80 (119.59 – 126.01)	123.81 (120.54 – 127.07)	0.67
Diastolic bp (mmHg)*	78.42 (75.89 – 80.95)	79.22 (76.69 – 81.74)	0.66

*Values are adjusted mean (95% CI)

Subjects Visit to the Clinical Site Facility: Baseline measures were obtained, including blood samples for laboratory analyses during Visit 1. The subjects again returned to the site on day 21 (visit 2) and day 42 (visit 3). All subjects made 3 visits to the clinical site. Prior to Visit 1 and coming to the site, subjects will complete a COVID pre-screen via telephone (e.g., vaccine status, current illness, etc.).

Project Compliance: An accurate and current accounting of the dispensing and return of study products for each subject was maintained on an ongoing basis by a member of the study site staff. The number of study products dispensed and returned by the subject was recorded on the Investigational Product Accountability Record. The study staff verified these documents throughout the course of the study.

Concomitant Medication: Records of medications prescribed during the clinical investigation to the enrolled subjects were maintained on the case report forms (CRFs). Reported medications included concomitant prescription medications, over-the-counter medications (OTC) and non-prescription medications.

Assessments of Efficacy: A subject's cognitive ability is generally evaluated using a variety of standardized questionnaires that included the Montreal Cognitive Assessment (MOCA®), the Mini Mental State Examination (MMSE), and the Adult ADHD Self-Reported Scale (ASRS-v1.1). All three assessments are well-validated instruments. While various cognitive aspects (executive functioning, visual short-term memory, language, working memory) were tested using games accessed from Happy Neuron Pro https://www.HappyNeuroPro.com/, a digital therapeutic tool. All these assessments were conducted in this investigation.

Primary Efficacy Endpoints

Montreal Cognitive Assessment (MOCA): It is validated as a highly sensitive tool for early detection of mild cognitive impairment (MCI) **[25]** The efficacy of GRE was assessed on MOCA at the initiation and at the end of 6 weeks of supplementation.

Mini Mental Scale Examination (MMSE): It is a set of 30-point questionnaires extensively used to assess cognitive impairment in patients, especially to determine their problems with thinking, communication, understanding and memory **[26]**. The efficacy of GRE was assessed on MMSE at the initiation and at the end of 6 weeks of supplementation.

Happy Neuron Cognitive Function Tests (HappyNeuroPro): It is a tool used by neurologists to help build the foundation of different cognitive functions that can ultimately lead to life improvement for the patient. Various cognitive aspects (executive functioning, visual short-term memory, language, working memory) were

tested using games accessed from https://www.HappyNeuroPro.com/, a digital therapy tool. A usual visit consisted of 4 different games, each played once, and in the same order: Game #155 (Ready, Steady, Count), Game #10 (Towers of Hanoi), Game #82 (Elephant Memory), and finally Game #17 (Shapes and colors). Games 155 (Ready, Steady, Count) and game 10 (Towers of Hanoi) were given two tries per session. Game #155 (Numbers Count Recall) involves working memory, executive function (calculation and arithmetic reasoning), mental imagery and subject's concentration. Game #10 (Executive Function Recall) determines problem-solving skills that call on the brain's executive functions–specifically planning and inhibition. In Game #82 (Verbal Memory), subjects were tested for their language and memory skills. Game #17 (Short-term Memory) assesses one's visual short-term memory. The efficacy of GRE was assessed on Games #155, #10, #82, and #17 at the initiation and end of 6 weeks of treatment.

Secondary Efficacy Endpoints

Adult ADHD Self Report Scale (ASRSv1.1):Adult ADHD Self-Report Scale (ASRS-v1.1) Symptom Checklist instructions (https://add.org/wp-content/uploads/2015/03/adhd-questionnaire-ASRS111.pdf) were used to determine the efficacy of GRE at the end of 6 weeks of treatment.

SF-36v1 Health Survey Quality of Life (QoL) & Global Evaluation Questionnaire: SF-36 is a set of generic, coherent, and easily administered quality-of-life measures. For the assessment of Quality of Life Health Survey, data was obtained from Visual Analog Scores of 0 to 100 evaluation presented to and filled by the participants. Physical Function, Role Physical Health, Role Emotional Health Energy/Fatigue, Emotional Well-Being, Social Functioning, Pain, and General Health were assessed at the baseline and at the completion of 6 weeks of treatment.

Global Evaluation Questionnaire: Subjects completed a Global Assessment Questionnaire on Visit 3 to evaluate their perceived efficacy of the treatment they received and whether they would purchase the product. A total of four questions were asked of each participant:

1. Do you believe the study product helped your memory?
2. Do you believe that the study product increased your attention span?
3. Do you believe that the product you were taking was: Active or Placebo
4. Would you purchase the study product if available to buy at a store or online?

Complete Comprehensive Metabolic Panel Laboratory Test/ Safety Assessment: For safety assessment, (a) Subject subjective adverse event form, (b) Vitals: heart rate (HR), systolic and diastolic blood pressure (BP), (c) Physical examination, (d) Medical history, and (e) Blood chemistries were assessed.

Safety of enrolled consuming placebo and investigational products was determined whenever the patient visited the clinical study site. Study participants visited the clinical study center three times, day 1, day 21, and day 42. The unused capsules, if any, were returned on the final day of the visit. Safety of the compound was assessed by critically monitoring for any adverse reaction during each follow-up visit and also by comparing the parameters at the baseline and on the last day of the visit.

Complete metabolic panel tests including serum glutamic oxaloacetic transaminase activity (SGOT/AST), serum glutamic pyruvic transaminase activity (SGPT/ALT), serum alkaline phosphatase activity (ALP), serum bilirubin, blood urea nitrogen (BUN) level, serum creatinine level, hemoglobin level, total leukocyte counts (TLC), serum urea, creatinine, levels were critically assessed at the baseline and at the completion of 6 weeks of supplementation.

Dropouts: Subjects dropped out from the study were critically monitored. The reasons for any dropouts were recorded.

Adverse Events Monitoring*:* Subjects were instructed to record all types of adverse events in their daily diaries during the duration of this investigation. Any major adverse events, subjects was provided a 24-hour emergency phone number to contact the principal investigator. Feedback from subjects were taken on whether they had incidences of any uncomfortable situation/problems or difficulties during their routine visits. Adverse event reporting was strictly ascertained.

Statistical Analyses: The statistical analysis was performed in a per-protocol fashion. For the primary outcomes associated with the validated instruments
(MMSE, MOCA, ASRS), and the continuous level laboratory parameters (sodium, potassium, glucose, etc.), ANCOVA (analysis of covariance) tests were performed with the treatment arms acting as the primary independent variable, the baseline observation of the continuous variable acting as a covariate, and the post-intervention observation of the continuous variable as the outcome/dependent variable. Marginal means with 95% confidence intervals were reported and interpreted for the ANCOVA analyses. Chi-square analysis was performed to compare the active and placebo treatment arms on the categorical outcomes measured in the GE assessment. Frequencies and percentages were reported and interpreted for the chi-square analyses. Independent samples t-tests and chi-square analysis were performed to compare the placebo and active treatment arms on demographic characteristics.

Paired t-tests were conducted to evaluate the parameters on the same individuals on Visit 1 (before administration of the Means and standard deviations were reported for the continuous demographic parameters and frequencies and percentage statistics were presented for the categorical demographic variables. All analyses were performed using SPSS Version 29 (Armonk, NY: IBM Corp.) and statistical significance was assumed at an alpha value of 0.05.

RESULTS

Study Participants*:* Initially, a total of 80 subjects (male = 45; female = 35; age = 20-55 years) participated in the study. At the end of the investigation, 5 subjects dropped out from the GRE group, while 7 subjects dropped out from the placebo group, while a total of 68 subjects (placebo = 33 subjects; GRE = 35 subjects) completed the study.

Cognitive Related Primary and Secondary Endpoints

Montreal Cognitive Assessment (MOCA), Mini Mental State Examination (MMSE), and Adult Attention Deficit Hyperactivity Disorder (ADHD) Self-Reported Scale (ASRS V1.1): Following six weeks of GRE supplementation, significant improvement was observed with ASRS (ASRS V1.1), however, no changes were observed with MOCA and MMSE.

Happy Neuron Pro Cognitive Function Tests (HappyNeuroPro): Various cognitive aspects (executive functioning, visual short-term memory, language, working memory) were assessed. A usual visit consisted of 4 different games, each played once, and in the same order: Game #155 (Ready, Steady, Count), Game #10 (Towers of Hanoi), Game #82 (Elephant Memory), and finally Game #17 (Shapes and colors). Games 155 (Ready, Steady, Count) and game 10 (Towers of Hanoi) were given to each participant two tries per session.

Table 3: Efficacy of GRE on MOCA, MMSE and ASRS Cognitive Assessments

Parameters	Placebo N = 33	GRE N = 35	*p*-value
MOCA*	26.39 (25.73 – 27.04)	26.29 (25.63 – 26.96)	0.84
MMSE*	28.12 (27.80-28.44)	28.11 (27.79 – 28.44)	0.97
ASRS*	38.73 (36.89 – 40.57)	35.14 (33.39-36.89	0.006**

*Values are adjusted mean (95% CI); **Statistically Significant, $p<0.0$

Table 4: Efficacy of GRE on Happy Neuron Pro Cognitive Function Tests

Parameters	Placebo N = 33	GRE N = 35	*p*-value
Game #155 Accuracy*	72.90 (60.57 – 85.24)	69.03 (54.13 – 83.93)	0.67
Game #155 Time*	22.81 (19.86 – 25.77)	20.19 (16.70 – 23.68)	0.24
Game #10 Accuracy*	31.39 (19.53 – 43.26)	47.16 (33.45 – 60.87)	0.09
Game #10 Time*	4.61 (4.22 – 5.00)	3.96 (3.50 – 4.41)	0.03**
Game #82 Accuracy*	80.00 (74.11 – 86.76)	87.67 (80.33 – 95.01)	0.14
Game #82 Time*	11.51 (8.59 – 14.43)	12.13 (8.56 – 15.70)	0.79
Game #17 Accuracy*	68.97 (61.33 – 76.62)	73.09 (64.25 – 81.93)	0.48
Game #17 Time*	1.64 (1.52 – 1.76)	1.50 (1.37 – 1.64)	0.13

*Values are adjusted mean (95% CI); **Statistically Significant, $p<0.05$

GAME #155 (Numbers Count Recall): As indicated earlier Game #155 involves working memory, executive function (calculation and arithmetic reasoning), mental imagery and your concentration. No significant difference was observed between the GRE-supplemented and the placebo groups for accuracy or time (p=0.67 and p=0.24, respectively).

GAME #10 (Executive Function Visual): As mentioned before Game #10 requires problem-solving skills that call on the brain's executive functions, specifically planning and inhibition. In Game #10, active subjects were much faster (time wise) than placebo in completing Game #10 (p = 0.03), which was statistically significant. However, there was a trend for GRE-supplemented subjects for their accuracy in the game, however it did not reach statistical significance (p=0.09).

GAME #82 (Verbal Memory): In Game #82, subjects assessed their language and memory skills. There was a trend for GRE supplemented subjects to have higher accuracy, however, it was not statistically significant (p=0.14). There was no significant difference with respect to time (p=0.79) between groups.

GAME #17 (Short-term Memory); Game #17 calls upon one's visual short-term memory. There was no difference between groups, but GRE supplemented subjects performed better for accuracy (p = 0.48), and faster for time (p=0.13).

It is important to emphasize that in Game #10 , which tests executive functioning, active subjects were much faster than placebo in completing the test (p = 0.03), which is statistically significant. There was also a trend in Active subjects for their accuracy, however, it did not reach statistical significance (p=0.09). For Game #82 (Verbal Memory), Active subjects had a trend for higher accuracy however it was statistically insignificant (p = 0.14). For Game #17 (Shapes and Colors), there was no difference between groups, but Active subjects performed better for accuracy (p = 0.48), and faster for time (p=0.13). These positive effects in all cases are quite promising and warrant further evaluation, as this indicates some effect of GRE.

SF-36V1 Quality of Life (QoL) Health Survey: For the QoL Health Survey, data was obtained from Visual Analog Scales (VAS) of 0 to 100 evaluation presented to and filled by subjects. Means and standard deviations of QoL questions are shown in Table 5, as changes from baseline, for GRE and placebo groups. Based on the linearly independent pairwise comparisons among the estimated marginal means and computed using alpha = 0.05, the active group scored significantly higher (p = 0.033) with respect to the parameters Role Physical Health, Role Emotional Problem, Energy/Fatigue, Emotional Well-Being and pain as compared to the placebo. The SF-36 results further asserted the beneficial role of GRE supplementation on the augmentation of cognitive abilities of the brain.

Variables	Placebo N = 33	GRE N = 35	p-value
Physical Function SF36*	94.69 (91.66 - 97.73)	94.79 (91.74 - 97.84)	0.97
Role Physical Health SF36*	88.68 (83.10 - 94.26)	95.32 (89.84 – 100.81)	0.09
Role Emotional Problem SF36*	81.62 (74.05 – 89.18)	93.09 (85.52 – 100.66)	0.033**
Energy/Fatigue SF36	63.80 (60.16 – 67.43)	63.99 (60.32 – 67.67)	0.94
Emotional Well-Being SF36	75.13 (71.84 – 78.42)	77.62 (74.27 – 80.97)	0.30
Social Functioning SF36	83.86 (80.05 – 87.67)	84.76 (81.03 – 88.49)	0.74
Pain SF36	90.44 (87.62 – 93.26)	88.84 (86.01 – 91.66)	0.42
General Health SF36	76.50 (73.37 – 79.63)	72.47 (69.34 – 75.60)	0.07

*Values are adjusted mean (95% CI); **Statistically Significant, $p<0.05$

Global Evaluation (GE) Questionnaire: As indicated earlier, all subjects completed a Global Assessment Questionnaire on Visit 3 to evaluate their perceived efficacy of the treatment. A total of four questions were asked of each subject, (Q1) whether GRE treatment helped your memory; (Q2) whether GRE boosted their attention span; (Q3) whether the consumed product was GRE or placebo; and (Q4) whether the participants will purchase GRE, if available to purchase at a store or online.

Subjects taking GRE supplementation were 1.47 times (p = 0.004; 95% CI 1.001 – 2.16) agreed that GRE boosted their memory and 1.96 times (p < 0.001; 95% CI 1.28 – 2.93) more likely in saying that GRE increased attention span. GRE-supplemented subjects were also

1.56 times (p = 0.02; 95% CI 1.05 – 2.32) more likely to purchase GRE, if it was available in store or one line.

Over 77% of GRE-supplemented subjects believed that the product increased their attention span and helped their memory by 70%. GRE-supplemented subjects were also more likely to purchase the product, as 70% of the GRE-supplemented group said they would buy the product if it was available commercially, either online or in-store. Figure 1 demonstrates the results.

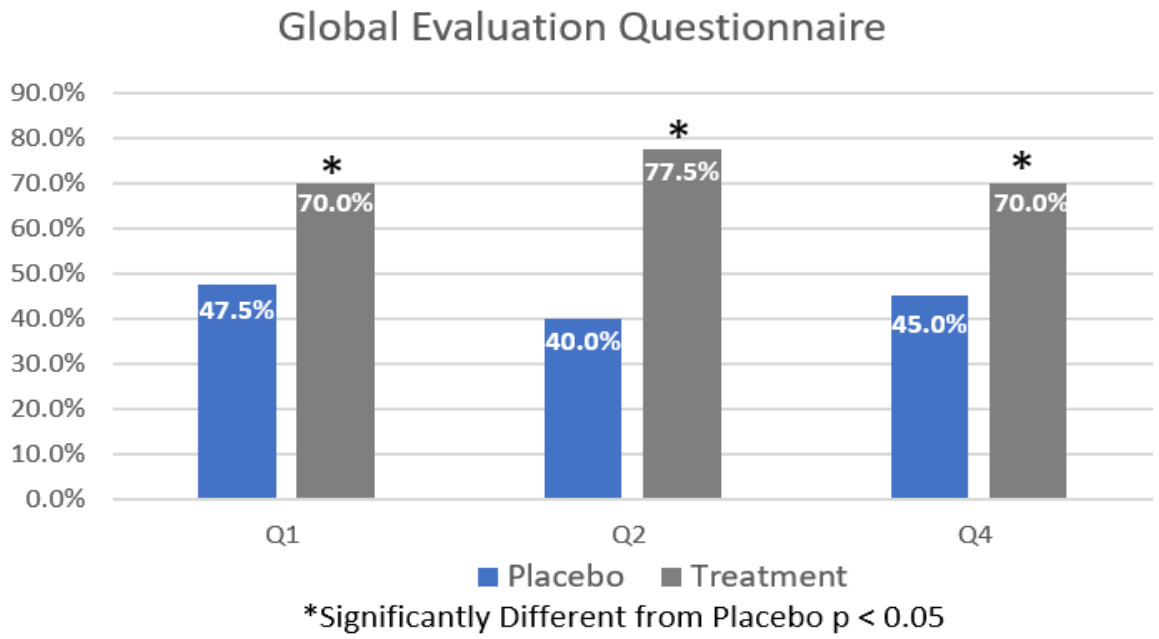


Figure 1: Global Health Questionnaire Findings

Complete Metabolic Panel (CMP) Laboratory Tests: After 6 weeks of GRE supplementation, reduction was noticed in the levels of Blood Urea Nitrogen (BUN)(Table 6), creatinine and SGPT/ALT levels. However, the values were within the normal clinical ranges. This confirmed that no untoward events were associated with the consumption of GRE.

Table 6: Selected Metabolic Parameters Data

Parameters	Placebo N = 33	GRE N = 35	p-value*	Reference Range Units
BUN*	13.27 (12.58 – 13.96)	11.98 (11.27 – 12.70)	0.011**	7 -20 mg/dL
BUN/Creatinine Ratio*	15.61 (14.70 – 16.53)	14.19 (13.27 – 15.10)	0.028**	3 – 40 Ratio
SGPT/ ALT*	28.09 (23.73 – 32.44)	18.17 (13.77 – 22.57)	<0.001**	< 40 U/L

Values are adjusted mean (95% CI). **Statistically Significant Finding, p < 0.05

Adverse Events: Some mild side effects were reported in this investigation. Five placebo and five GRE treated subjects reported temporary side effects including nausea, headache, stomach pain, diarrhea, constipation, and vivid dreams. Additionally, one female subject in the treatment group reported that her menstrual cycle off by

dates and her skin felt dry. However, all these side effects were quite mild and temporary for both placebo and GRE-supplemented groups.

Dropouts: At the end of the clinical investigation, 5 subjects dropped out from the GRE group, while 7 subjects dropped out from the placebo group. Thus, a total of 68 subjects successfully completed the clinical trial.

DISCUSSION

Brain utilizes approximately 20 percent of the total basal oxygen turnover of the body [27] and therefore, the brain tissues, with a prolific rate of oxidative phosphorylation in the neuronal mitochondria, are much more prone to ROS-induced damage than any other organ or tissue of the body. Oxidative stress inflicts damage to the neurons via two broad mechanisms, a) the reactive species of oxygen and nitrogen oxidize cellular macromolecules leading to cell death b) it also initiates downstream redox signaling pathways including the nuclear factor (NF)-κB, mitogen-activated protein kinases (MAPK), the phosphoinositide 3-kinase (PI3K)/Akt pathway, all of which culminate towards the generation of inflammatory response, cell death by apoptosis and onset of neurodegenerative disorders [28,29]. Lipid peroxidation, particularly, poses a serious concern to brain cells owing to their high content of oxidizable lipid and metal ions [30]. Under the circumstances, the use of potent antioxidants to sequester and scavenge free radicals has emerged to be the most promising therapeutic option. In this regard, Nitric Oxide Synthase (NOX) inhibitors, NMDA receptor blockers, and other Non-Steroidal Anti Inflammatory Agents (NSAIDs) are useful therapeutics that can mitigate oxidative damage in brain cells [31].

Despite significant advances toward understanding oxidative stress-induced damage in brain cells, a combinatorial antioxidant therapy has not come out in the market which can safeguard us against sudden brain strokes or the development of critical neurodegenerative disorders, probably due to the generation of an excess of ROS that sets off an uncontrollable avalanche of deleterious effects inside the cell. In this respect, the anti-oxidative, anti-inflammatory, and anti-cholinesterase activities of plant-derived tannins have demonstrated neuroprotective activity against neurodegenerative as well as neuropsychiatric disorders [32].

In the present study, the effect of gallotannins from Sumac gall on the improvement of cognitive functions was investigated by a combination of primary and secondary endpoint tools for investigating any cognitive problem. The Mini-Mental State Exam (MMSE) is widely used in detecting early symptoms of Alzheimer's disease and includes components such as orientation capabilities, word recalling power, comprehension of language, attention span, ability to calculate and execute visuospatial analysis. The Montreal Cognitive Assessment (MoCA) is a newer and more refined tool that incorporates additional tools such as clock drawing and connecting dots [33]. A significant reduction was observed in Adult ADHD Self-Reported Scale (ASRS). Subsequently, the patients were subjected to HappyNeuronPro, a collective set of web-based cognitive assessment tools. HappyNeuronPro has a number of cognitive therapies exercises that target areas of Executive Functioning, Verbal Memory, Visual Memory, Spatial Memory, Visual and Spatial Abilities, Visual Attention, Processing Speed, and Auditory areas. Exercises include deciphering quotations, sorting numbers in a grid, mentally rotating figures, categorizing words, and determining where to place them [34]. A significant reduction in time was obtained only with respect to Game 10 which suggested that there was marked improvement in processing in the frontal region of the brain with respect to taking fast decisions and using a logical problem-solving approach. Subsequently,

the patients were subjected to the 36-Item Short Form Health Survey questionnaire (SF-36) tool, a popular and fast-emerging technique for assessing the quality of life about health [35]. A significant increase was observed with respect to the Role Emotional Problem comprising three items. This also indicated that on consumption of GRE, emotional health was also improved significantly. Lastly, analysis of the Global Evaluation (GE) Questionnaire strongly supported the beneficial role of gallotannins in substantially improving the overall functioning of the brain as more than 70% of patients reported significant improvement in their cognitive functions and were willing to purchase the therapeutic again if available.

A preliminary study was conducted by Woo and Kee (unpublished data) to unveil the molecular mechanism of GRE that had provided useful insights to understand its protective abilities against oxidative stress-induced neuronal damage. It was observed that administration of GRE resulted in substantial upregulation of Sigma 1 receptor mRNA. However, these findings need to be reconfirmed by conducting additional studies. The Sigma 1 receptors are a group of multi-functional receptors present in the endoplasmic reticulum of neurons. Gross changes in the expression of Sigma 1 are associated with the onset of different neurodegenerative disorders including Alzheimer's and Parkinson's Disease [36]. Under normal cellular conditions, the Sig-1R is bound to the chaperone-binding immunoglobulin protein (BiP)/glucose-regulated protein 78 (GRP78) and together remains docked at the mitochondria-associated ER membrane (MAM). Upon stress induction, Sig-1R gets dissociated from Bip and docks with type 3 inositol 1,4,5-trisphosphate receptor3 (IP3R3) at other regions including the plasma membrane, the ER membrane, and the nuclear envelope [37] from where it mediates interactions with a plethora of ion channels and receptors implicated in calcium signaling pathway [38] and recruits many chromatin remodeling factors [39]. GRE induced upregulation in the expression of Sigma receptor indicated that the CNS was better equipped to thwart any neuronal malfunction arising out of oxidative damage.

Oxidative damage causes misfolding and aggregation of proteins resulting in the development of amyloid aggregates [40]. Amyloid-like aggregation has also been reported for glycation-induced aggregation of proteins thus linking the pathophysiology of diabetes with neurodegenerative disorders [41,42]. Administration of GRE may be able to curtail the accumulation of amyloid aggregates or Lewy bodies by as much as 70% in a hydrogen peroxide-induced oxidative stress model of mouse brain which confirmed that GRE was able to improve the cognitive health by reversing the effects of oxidative stress and concomitant accumulation of free radicals (unpublished data). However, we need to reconfirm the data by conducting additional studies.

CONCLUSION

Brain stress and brain stroke arising from oxidative stress is one of the significant pathophysiologies leading to mental stress, memory loss, fatigue and even possibly more serious issues like dementia, Alzheimer's disease, and other debilitating neurodegenerative ailments. Originally a hallmark of aging, more and more incidences of chronic neuronal damage leading to severe cases cognitive impairment are coming to the forefront mostly arising out of stress associated with modern-day lifestyle. Although the major molecular crosstalk's implicated in ROS-induced inflammatory response and other signaling pathways involved in the normal functioning of the neurons in some diseases have been largely delineated [43], we are yet to find out an effective combination of antioxidants or related phytonutrients to effectively arrest, slow or reverse the damage caused by the reactive oxygen species (ROS) produced in abundance in the

neuronal mitochondria. In this respect, plant-derived alkaloids such as Gallotannins present in the proprietary Galla Rhois Extract (GRE) in our study exhibited the ability to boost neuronal well-being in young and aged people. Therefore, it is prospective phytotherapeutic to manage ROS induced damage especially to brain tissues. However, further success of the formulation will depend on unraveling the molecular targets of GRE to design novel therapeutic intervention strategiesMoreover, this extract can also be incorporated in functional foods or in brain boosting supplements to achieve effective augmentation of cognitive performance in them,. Additional and larger studies are being planned to further elucidate and substantiate these findings and mechanisms.

Abbreviations: ADHD: Attention Deficit Hyperactivity Disorder; ASRSv1.1: Adult ADHD Self-Reported Scale; ALP: Alkaline Phosphatase; ALT: Alanine Transaminase; AST: Aspartate Aminotransferase; BUN: Blood Urea Nitrogen; CMP: Complete Metabolic Panel; GE: Global Evaluation; GRE: Galla Rhois Extract; IRB: Institutional Review Board; MOCA: Montreal Cognitive Assessment; MMSE: Mini Mental Scale Examination; ROS: Reactive Oxygen Species; SF-36 Tool: 36-Item Short Form Health Survey questionnaire; SGOT: Serum Glutamic Oxaloacetic Transaminase Activity; SGPT: Serum Glutamic Pyruvate Transaminase; MAM: Mitochondria Associated ER Membrane; QoL: Quality of Life; TLC: Total Leukocyte Counts.

Author's contributions: All authors contributed equally and reviewed the version of the manuscript to be submitted.

Competing Interests: The authors declare no competing interest.

Acknowledgements: All authors thank Global Clinicals, Inc., Los Angeles, CA, (www.globalclinicals.com), for meticulously conducting the clinical study. Also, the authors appreciate and thank R. Eric Heidel, PhD, PStat, Associate Professor of Biostatistics, University of Tennessee Graduate School of Medicine, for performing the statistical analysis of this investigation.

REFERENCES

1. Singh, A., Kukreti, R., Saso, L., and Kukreti, S.: Oxidative stress: A key modulator in neurodegenerative diseases. Molecules. 2019, 24(8):1583. DOI: https://doi.org/10.3390/molecules24081583.
2. Rink, C., and Khanna, S.: Significance of brain tissue oxygenation and the arachidonic acid cascade in stroke. Antioxid Redox Signal. 2011, 14(10):1889-1903. DOI: https://doi.org/10.1089/ars.2010.3474.
3. Bruce, K.D., Zsombok, A., and Eckel, R.H.: Lipid processing in the brain: A key regulator of systemic metabolism. Front Endocrinol (Lausanne). 2017, 8:60. DOI: https://doi.org/10.3389/fendo.2017.00060.
4. Suzuki, N., Sawada, K., Takahashi, I., Matsuda, M., Fukui, S., Tokuyasu, H., Shimizu, H., Yokoyama, J., Akaike, A., and Nakaji, S.: Association between polyunsaturated fatty acid and reactive oxygen species production of neutrophils in the general population. Nutrients. 2020, 12(11):3222. DOI: https://doi.org/10.3390/nu12113222.
5. Salim S.: Oxidative stress and the central nervous system. J Pharmacol Exp Ther. 2017, 360(1):201-205. DOI: https://doi.org/10.1124/jpet.116.237503.
6. Puspita, L., Chung, S.Y., and Shim, J.W.: Oxidative stress and cellular pathologies in Parkinson's disease. Molecular Brain. 2017, 10(1):53. DOI: https://doi.org/10.1186/s13041-017-0340-9.
7. Aman, Y., Ryan, B., Torsetnes, S.B., Knapskog, A.B., Watne, L.O., McEwan, W.A., and Fang. E.F.: Enhancing mitophagy as a therapeutic approach for neurodegenerative diseases. Int Rev Neurobiol. 2020, 155:169-202. DOI: https://doi.org/10.1016/bs.irn.2020.02.008.
8. Wan, J., Ren, H., and Wang, J.: Iron toxicity, lipid peroxidation and ferroptosis after intracerebral

hemorrhage. Stroke and Vascular Neurology 2019, 4(2) DOI: https://doi.org/10.1136/svn-2018-000205.

9. Jiang, X., Stockwell, B.R., and Conrad, M.: Ferroptosis: mechanisms, biology and role in disease. Nat Rev Mol Cell Biol. 2021, 22(4):266–282. DOI: https://doi.org/10.1038/s41580-020-00324-8.
10. Sienes Bailo, P., Llorente, M, Elena, C., Pilar, M.B., Silvia, B.G., Adrián, P.G., Adela, S.P.C., Joan, J., Vaquer, S., Juana, M., Dayaldasani Khialani, A., Cedra, M., Concepcion, C.A., Guillermo, S.T.J., and Fort Gallifa, I.: The role of oxidative stress in neurodegenerative diseases and potential antioxidant therapies. Advances in Laboratory Medicine / Advances en Medicina de Laboratorio 2022, 3(4): 342-350. DOI: https://doi.org/10.1515/almed-2022-0111
11. He, H.F.: Recognition of Gallotannins and the Physiological Activities: From Chemical View. Front Nutr. 2022 Jun 1; 9:888892. DOI: https://doi.org/10.3389/fnut.2022.888892. PMID: 35719149; PMCID: PMC9198600.
12. Zhang, Y., Wang, M., and Chang, W.: Iron dyshomeostasis and ferroptosis in Alzheimer's disease: Molecular mechanisms of cell death and novel therapeutic drugs and targets for AD. Front Pharmacol. 2022 Sep 16; 13:983623. DOI: https://doi.org/10.3389/fphar.2022.983623.
13. Jelinek, M., Jurajda, M., and Duris, K.: Oxidative Stress in the Brain: Basic Concepts and Treatment Strategies in Stroke. Antioxidants (Basel). 2021, 10(12):1886. DOI: https://doi.org/10.3390/antiox10121886.
14. Limanaqi, F., Biagioni, F., Mastroiacovo, F., Polzella, M., Lazzeri, G., and Fornai, F.: Merging the Multi-Target Effects of Phytochemicals in Neurodegeneration: From Oxidative Stress to Protein Aggregation and Inflammation. Antioxidants (Basel). 2020, 9(10):1022. DOI: https://doi.org/10.3390/antiox9101022.
15. Rebas, E., Rzajew, J., Radzik, T., and Zylinska, L.: Neuroprotective polyphenols: A modulatory action on neurotransmitter pathways. Curr. Neuropharmacol. 2020, 18:431–445. DOI: https://doi.org/10.2174/1570159X18666200106155127
16. Bordoni, L., and Gabbianelli, R.: Mitochondrial DNA and neurodegeneration: Any role for dietary antioxidants? Antioxidants. 2020, 9:764. DOI: https://doi.org/10.3390/antiox9080764.
17. Naoi, M., Wu, Y., Shamoto-Nagai, M., and Maruyama, W.: Mitochondria in neuroprotection by phytochemicals: Bioactive polyphenols modulate mitochondrial apoptosis system, function and structure. Int J Mol Sci. 2019, 20(10):2451. DOI: https://doi.org/10.3390/ijms20102451.
18. Park, J.W., Kim, J.E., Kang, M.J., Choi, H.J., Bae, S.J., Kim, S.H., Jung, Y.S., Hong, J.T., and Hwang, D.Y.: Anti-oxidant activity of Gallotannin-enriched extract of Galla Rhois can associate with the protection of the cognitive impairment through the regulation of BDNF signaling pathway and neuronal cell function in the scopolamine-treated ICR mice. Antioxidants (Basel). 2019 Oct3;8(10):450. DOI: https://doi.org/10.3390/antiox8100450.
19. Al-Halabi, R., Bou Chedid, M., Abou Merhi, R., El-Hajj, H., Zahr, H., Schneider-Stock, R., Bazarbachi, A., and Gali-Muhtasib, H.: Gallotannin inhibits NFκB signaling and growth of human colon cancer xenografts. Cancer Biol Ther. 2011, 12(1):59-68. DOI: https://doi.org/10.4161/cbt.12.1.1571.
20. Kim, S.H., Jun, C.D., Suk, K., Choi, B.J., Lim, H., Park, S., Lee, S.H., Shin, H.Y., Kim, D.K., and Shin, T.Y.: Gallic acid inhibits histamine release and pro-inflammatory cytokine production in mast cells. Toxicol Sci. 2006, 91:123–31. DOI: https://doi.org/10.1093/toxsci/kfj063.
21. Lee, M.K., Y.H. Hwang, H. Ryu, A. Lee, H.H. Jeong, J. Baek, M.J. Kim MJ, J.Y. Lee, J.Y. Van, Y. Liu, C.W. Choi, M.S. Kim, and B. Lee. Galla rhois water extract inhibits enzymatic browning in apple juice partly by binding to and inactivating polyphenol oxidase. Food Chem. 2022, 383:132277. DOI: https://doi.org/10.1016/j.foodchem.2022.132277.
22. Lee, Y.G., Kang, K.W., Hong, W., Kim, Y.H., Oh, J.T., Park, D.W., Ko, M., Bai, Y.F., Seo, Y.J., Lee, S.M., Kim, H., and Kang, S.C.: Potent antiviral activity of Agrimonia pilosa, Galla rhois, and their components against SARS-CoV-2. Bioorg Med Chem. 2021, 45:116329. DOI: https://doi.org/10.1016/j.bmc.2021.116329.
23. Lee, K., Kim, J., Lee, B.J., Park, J.W., Leem, K.H., and Bu, Y.: Protective effects of Galla Rhois, the excrescence produced by the sumac aphid, Schlechtendalia chinensis, on transient focal cerebral ischemia in the rat. J Insect Sci. 2012, 12:10. DOI: https://doi.org/10.1673/031.012.0110.
24. Sylla, T., Pouységu, L., Da Costa, G., Deffieux, D., Monti, J.P., and Quideau, S.: Gallotannins and tannic acid: First chemical syntheses and in vitro inhibitory activity on Alzheimer and amyloid β-peptide aggregation. Angew Chem Int Ed Engl. 2015, 54(28):8217-21. DOI: https://doi.org/10.1002/anie.201411606.
25. O'Driscoll, C., and Shaikh, M.: Cross-Cultural Applicability of the Montreal Cognitive Assessment (MoCA): A Systematic

Review. J Alzheimers Dis. 2017, 58(3):789-801. DOI: https://doi.org/10.3233/JAD-161042.

26. Arevalo-Rodriguez, I, Smailagic, N., Roqué-Figuls, M., Ciapponi, A., Sanchez-Perez, E., Giannakou, A., Pedraza, O.L., Bonfill Cosp, X., and Cullum, S.: Mini-Mental State Examination (MMSE) for the early detection of dementia in people with mild cognitive impairment (MCI). Cochrane Database Syst Rev. 2021, 7(7):CD010783. DOI: https://doi.org/10.1002/14651858.CD010783.pub3.
27. Cobley, J.N., Fiorello, M.L., and Bailey, D.M.: 13 reasons why the brain is susceptible to oxidative stress. Redox Biol. 2018, 15: 490-503. DOI: https://doi.org/10.1016/j.redox.2018.01.008.
28. Dong, H., Zhang, Y., Huang, Y., and Deng, H.: Pathophysiology of RAGE in inflammatory diseases. Front Immunol. 2022, 13:931473. DOI: https://doi.org/10.3389/fimmu.2022.931473.
29. Sekowski, S., Olchowik-Grabarek, E., Dubis, A.T., Sharan, L., Kumar, A., Abdulladjanova, N., Markiewicz, P., and Zamaraeva, M.: Inhibition of AGEs formation, antioxidative, and cytoprotective activity of Sumac (Rhus typhina L.) tannin under hyperglycemia: molecular and cellular study. Mol Cell Biochem. 2023, 478(3):443-457. DOI: https://doi.org/10.1007/s11010-022-04522-0.
30. Júnior, H.V., de França Fonteles, M.M., and Mendes de Freitas, R. Acute seizure activity promotes lipid peroxidation, increased nitrite levels and adaptive pathways against oxidative stress in the frontal cortex and striatum. Oxid Med Cell Longev. 2009, 2(3):130-137. DOI: https://doi.org/10.4161/oxim.2.3.8488.
31. Nguyen, A., Patel, A.P., Kioutchoukova, I.P., Diaz, M.J., and Lucke-Wold, B.: Mechanisms of mitochondrial oxidative stress in brain injury: From pathophysiology to therapeutics. Oxygen 2023, 3, 163-178. DOI: https://doi.org/10.3390/oxygen3020012.
32. Hussain, G., Huang, J., Rasul, A., Anwar, H., Imran, A., Maqbool, J., Razzaq, A., Aziz, N., Makhdoom, E.U.H., Konuk, M. and Sun, T.: Putative roles of plant-derived tannins in neurodegenerative and neuropsychiatry disorders: An updated review. Molecules. 2019, 24(12):2213. DOI: https://doi.org/10.3390/molecules24122213.
33. Dautzenberg, G., Lijmer, J. and Beekman, A.: Diagnostic accuracy of the Montreal Cognitive Assessment (MoCA) for cognitive screening in old age psychiatry: Determining cutoff scores in clinical practice. Avoiding spectrum bias caused by healthy controls. Int J Geriatr Psychiatry. 2020, 35(3):261-9. DOI: https://doi.org/10.1002/gps.5227.
34. Brevik, E.J., Lundervold, A.J., Haavik, J. and Posserud, M.B.: Validity and accuracy of the Adult Attention-Deficit/Hyperactivity Disorder (ADHD) Self-Report Scale (ASRS) and the Wender Utah Rating Scale (WURS) symptom checklists in discriminating between adults with and without ADHD. Brain Behav. 2020, 10(6): e01605. DOI: https://doi.org/10.1002/brb3.1605. Epub 2020 Apr 13. Erratum in: Brain Behav. 2021, 11(5): e02067. PMID: 32285644; PMCID: PMC7303368.
35. Lins, L., Carvalho, F.M.: SF-36 total score as a single measure of health-related quality of life: Scoping review. SAGE Open Med. 2016, 4:2050312116671725. DOI: https://doi.org/10.1177/2050312116671725.
36. Ryskamp, D.A., Korban, S., Zhemkov, V., Kraskovskaya, N., and Bezprozvanny, I.: Neuronal sigma-1 receptors: Signaling functions and protective roles in neurodegenerative diseases. Front Neurosci. 2019, 13:862. DOI: https://doi.org/10.3389/fnins.2019.00862.
37. Hayashi, T., and Su, T.P.: Sigma-1 receptor chaperones at the ER-mitochondrion interface regulate Ca(2+) signaling and cell survival. Cell. 2007, 131(3):596-610. DOI: https://doi.org/10.1016/j.cell.2007.08.036.
38. Su, T.P, Hayashi, T., Maurice, T., Buch, S. and Ruoho, A.E.: The sigma-1 receptor chaperone as an inter-organelle signaling modulator. Trends Pharmacol Sci. 2010, 31(12):557-566. DOI: https://doi.org/10.1016/j.tips.2010.08.007.
39. Tsai, S.Y., Chuang, J.Y. Tsai, M.S., Wang, X.F., Xi, Z.X., Hung, J.J., Chang, W.C., Bonci, A. and Su, T.P.: Sigma-1 receptor mediates cocaine-induced transcriptional regulation by recruiting chromatin-remodeling factors at the nuclear envelope. Proc Natl Acad Sci U S A. 2015, 112(47):E6562-6570. DOI: https://doi.org/10.1073/pnas.1518894112.
40. Zhao, Y., and Zhao, B.: Oxidative Stress and the Pathogenesis of Alzheimer's Disease. Oxid Med Cell Longev. 2013; 2013:316523. DOI: https://doi.org/10.1155/2013/316523.
41. Das, A., Basak, P., Pramanik, A., Majumder, R., Ghosh, A., Hazra, S., Guria, M., Bhattacharyya, M. and Banik, S.P.: Ribosylation induced structural changes in Bovine Serum Albumin: understanding high dietary sugar induced protein aggregation and amyloid formation. Heliyon. 2020, 6(9):e05053. DOI:

https://doi.org/10.1016/j.heliyon.2020.e05053.

42. Sirangelo, I. and Iannuzzi, C.: Understanding the Role of Protein Glycation in the Amyloid Aggregation Process. Int J Mol Sci. 2021, 22(12):6609. DOI: https://doi.org/10.3390/ijms22126609.
43. Vijayan, M., and Reddy, P.H.: Stroke, vascular dementia, and Alzheimer's disease: Molecular links. J Alzheimers Dis. 2016, 54(2):427-443. DOI: https://doi.org/10.3233/JAD-160527.