## Original Article

# Holy basil (Ocimum sanctum Linn.) leaf extract enhances specific cognitive parameters in healthy adult volunteers: A placebo controlled study

**Suneetha Sampath[1], S.C. Mahapatra[1], M.M. Padhi[2], Ratna Sharma[1] and Anjana Talwar*[1]**

[1] Department of Physiology,
  All India Institute of Medical Sciences,
  New Delhi, India
[2] Central Council for Research in Ayurveda and Siddha,
  Department of Ayurveda, Yoga and Naturopathy, Unani and Siddha (AYUSH),
  New Delhi, India

## Abstract

**Introduction:** *Ocimum sanctum* (OS), known as Holy basil, has been documented to possess neuro-protective, cognition-enhancing and stress relieving effects in animal models. However there is paucity of clinical studies to document these effects.

**Materials and methods:** Effect of OS on parameters related to cognition and stress in humans was evaluated with administration of 300 milligram capsules of ethanolic leaf extracts of *Ocimum sanctum* (EtOS) or placebo per day, over 30 days.

**Results:** Intra-group comparison of Sternberg and Stroop test showed improvement in both the placebo and EtOS groups, however, the improvement stabilized after day 15 in the placebo group. Intergroup comparison revealed a significant improvement of the following cognitive parameters in the EtOS as compared to the placebo: reaction time (RT) and error rate (ER) of Sternberg test, RT of neutral task of Stroop, RT and ER of interference task of Stroop.

The intra-group comparison of P300 latency, salivary cortisol, and State-Trait Anxiety Inventory showed improvement over time in the EtOS group alone, though the inter-group difference was significant in the P300 latency alone. There were no changes in heart rate (HR), ΔHR, or galvanic skin response (GSR) or ΔGSR.

**Conclusion**: *Ocimum sanctum* leaf extract seems to have potential cognition-enhancing properties in humans.

## Introduction

Ocimum tenuiflorum, also known as *Ocimum sanctum* (OS) (Holy Basil) is an aromatic plant in the family Lamiaceae, native to south-east Asia and the tropics. It is referred to as 'Tulsi' in India, and has been used extensively in ayurveda, the traditional Indian system of medicine. Extracts of the leaves of OS are known for its calming and relaxing effects. Alcoholic extract of leaves of OS contain ursolic acid, apigenin, luteolin, apigenin-7-O-glucuronide, luteolin-

**\*Corresponding author :**
Dr. Anjana Talwar, Dept of Physiology, All India Institute of Medical Sciences, New Delhi, India
Email: anjanatalwar@gmail.com
(Received on September 10, 2014)

7-O-glucuronide, isorientin, orientin, molludistin, stigmasterol, vicenin-2, vitexin, isovitexin, aesculetin, aesculin, chlorgenic acid, galuteolin, circineol, gallic acid, procatechuic acid, vallinin acid, chlorogenic acid, β stigmasterol (1).

Preliminary evaluation of the psychopharmacological effects of EtOS showed that it had anti-convulsant, anti-anxiety and anti-depressant effects (2).

Administration of Ethanolic extract of OS (EtOS) had a favorable effect on cognitive parameters in animal models. EtOS ameliorated the amnesic effects in cognitive dysfunction in rat model (3, 4) and improved learning and memory in alzheimer's rat model (5). EtOS also had beneficial effects in cerebral reperfusion injury and cerebro-vascular insufficiency states (6, 7).

Treatment with EtOS normalized the alterations in various parameters like plasma corticosterone (8), WBC count and neutrophil function (9) induced by acute and chronic noise stress, in albino rats. High doses of EtOS also had a nervous system stimulant and/or antistress effects comparable to that of desipramine, as assessed by mouse swimming performance test (10). OS containing formulations (EuMil) attenuated chronic stress induced alterations in glucose intolerance, suppression of male sexual behavior, and cognitive dysfunction in albino rats, in a dose dependent manner, comparable to that of Ginseng (11). Aqueous extracts of OS and C. sinensis reduced the behavioral alterations induced by restraint stress in rats (12). EtOS showed anti-anxiety and anti-depressant effects, as evaluated by various behavioral experiments (13).

The psychopharmacological effects of OS have been postulated to involve dopaminergic neurons as the effects were blocked by haloperidol, and synergized by bromocriptine (2). Ocimumosides A, B and 4-allyl-1-O-beta-D-glucopyronosyl-2-hydroxybenzene have been postulated for the the anti-stress properties of OS (14). The abundance of phenolics and flavanoids have been held responsible for the anti-oxidant effect of EtOS (15).

Formulations containing extracts of OS, ameliorated the chronic stress-induced neurochemical perturbations in rats. There was a normalization of the concentrations of Noradrenaline (NA) and dopamine (DA) in frontal cortex, pons-medulla, hypothalamus hippocampus and that of 5-hydroxytryptamine (5HT) in frontal cortex, pons medulla, hypothalamus and hippocampus, induced by electroshock stress (16). Administration of 70% EtOS also provided similar results, normalizing the neurotransmitter levels in discrete brain areas induced by noise stress (17, 18).

EtOS normalized the reduction in total acetylcholine content and the increase in the activity of acetylcholinesterase in cerebral cortex, corpus striatum, hypothalamus and hippocampus of rat brain (19, 4). EtOS also attenuated the lipid peroxidation by normalizing the levels of superoxide dismutase , catalase, glutathione peroxidase and glutathione, in a dose-dependent manner (15). EtOS also possessed strong free radical scavenging action and provided increased resistance against thermal stress in C. elegans which has been postulated due to modulation of some signaling pathways (20).

Despite availability of extensive pre-clinical evidences on anti-stress activity of OS, only two clinical reports have documented beneficial effects of OS in questionnaire based study. OS extract 500mg twice a day, taken over 60 days, significantly attenuated stress and depression levels in 35 patients with generalized anxiety disorder (16). Intake of OciBest®, a commercial preparation of OS, at dose of 1200 mg/day over 6 weeks, resulted in symptomatic improvement based on a self-reported questionnaire in 71 patients having symptoms of stress (21). Dose of Ocimum Sanctum used in these studies was higher in comparison to the dose used (300 mg/day) for eliciting the Immuno-modulatory effects of EtOS in healthy subjects in our laboratory (22). In these subjects there was an increase in the levels of IFN-γ, IL-4 and percentages of T-helper cells and NK-cells after 4 weeks of intake of EtOS. Thus, in the present study also, EtOS was used in the dose of 300 mg/day for thirty days.

The study was designed to investigate the cognition enhancing and anti-stress potential of ethanolic leaf

extract of *Ocimum Sanctum.*

## Participants and methods

### Study design

The study was performed in a double-blinded randomized control manner after obtaining ethical clearance from institutional ethics committee and registered with clinical trial registry of India (CTRI) CTRI/2010/091/006113. The study was conducted at the Deptt. of Physiology, AIIMS, New Delhi.

### Subjects

44 apparently healthy male subjects, aged 18-30 years, were recruited on voluntary basis. Subjects suffering from long term diseases/disabilities, chronic smokers and those on any long term medication were excluded from the study based on a clinical interview. Subjects were asked to abstain from smoking, alcohol and caffeine containing beverages on the day of the experimental session.

On the day before the first recording session, subjects were briefed about the study, familiarized with the recording procedures. A written informed consent was obtained and subjects were asked to pick up a lottery and randomly allocated into either group A or B. Recordings were done on day 0 (D0), day 15 (D15) and day 30 (D30). 23 subjects were recruited in group A and 21 in group B.

### Interventional drug (*Ocimum sanctum* extract/placebo):

70% ethanolic extract of *Ocimum sanctum* (EtOS), prepared from dried leaves, was supplied by central council for research in ayurveda and siddha (CCRAS), India for which the leaves of *Ocimum sanctum L.* were procured from Dindigal area of Tamil Nadu, South India in October 2006 and its authenticity and the identity was established by careful scrutiny of taxonomical features after the consultation of literature *Flora of Presidency of Madras.* Vol. 2 Adlard & Sons Ltd. and Database on Medicinal Plants used in Ayurveda, Vol.2 by CCRAS, New Delhi, and after matching with the raw material specimen R.D. no 792 in Raw Drug Museum of CCRAS identified as

*Ocimum sanctum* L. leaf, it was deposited in raw drug museum of CCRAS under the raw drug serial No 6001 for further reference.

Quality assessment run for EtOS was done by Quality Control Laboratory, M/s. chemiloids, Laila Impex, Vijayawada, India lab reference no.L 7040426, for solvent residues, heavy metals, pesticide residues, microbiological contamination and phyto-chemical specifications (ursolic acid >2.7% w/w). Placebo (sucrose) was supplied by the Dabur Pharmaceutical (India) Ltd., Ghaziabad (U.P.), India. Capsules of 300 milligram of either the drug or placebo (sucrose) were identically capsulated and could not be distinguished from each other. 30 capsules of EtOS or placebo were packaged in identical air-sealed bottles and labelled A or B, by the GLP certified laboratory of Dabur research foundation, India. The packed containers were dispatched from laboratory of Dabur research foundation, India to the study site. The subject and the experimenter were blinded to the identity of capsules in the bottles.

Immuno-modulatory effects of EtOS in healthy subjects have been documented using the same capsules at the same dose (300 mg/day), in our lab, and no adverse effects were reported (22). Subjects in group A and B took one capsule a day from bottle A or B respectively, each morning, on empty stomach. They were instructed to record and report any health related symptom during this period, in the absence of which, they reported back for the assessment and recording on D15 and D30.

### Assessment of cognitive function:

Sternberg and Stroop task were administered through computerized Psych/Lab™ Software (Richard A Abrahams, Department of Psychology, Washington, University of Missouri, USA). Event related potential (ERP) was assessed using RMS EMG EP-Mark II system, Recorders and Medicare Systems Pvt Ltd, Chandigarh, India.

Sternberg memory task tests the accuracy and speed of short term memory load. The program 'MEMSCAN' runs a version of the Sternberg memory scanning paradigm. There were 3 blocks containing 12 trials

each (1st block not taken for analysis). On each trial, a set of to-be-remembered digits was first presented. The size of the set varies on each trials from 1 to 6. The memory set is then replaced by a plus sign and, after a short delay, a probe digit. The subject's task is to respond as quickly as possible by pushing the forward slash key if the probe was a member of the set, or the Z key if the probe was not a member of the memory set. The reaction time (RT) and error rate (ER) were measured (23).

The Stroop test measures cognitive flexibility. The program 'STROOP' runs a version of the Stroop paradigm. The Stroop test measures the ease with which a subject can shift his perception to conform to changing demands and inhibit attention to competing stimuli. The subject has to respond to the color in which the word is printed by pressing the appropriate key as quickly as possible. Three different paradigms of facilitation, neutral and interference tasks were administered in separate blocks, each with 24 trials. In neutral task the letter string is composed of Xs. In interference task the letter string is one of the words red, green, blue, or yellow printed in a color different from the named color. In facilitation task the letter string is the name of the color that the letters are printed in. On each trial, the subjects were presented with a string of letters printed in color; and they were instructed to respond to the colour in which the word was printed, by pressing the appropriate colored key of the keyboard as quickly as possible (24).

Auditory event-related potentials were elicited using auditory oddball paradigm, with amplitude, 50-60 dB greater than the auditory threshold of the subject. The standard (1000 Hz) and the target tone (2000 Hz) were in the frequency of 4:1 and the results of 36 rare stimuli were averaged.

The event related potential was recorded using the EEG activity (filter bandpass:0.1-50Hz, analysis time:1sec) recorded at Cz and Pz sites according to the 10/20 system. The resistance was kept below 5 kohm. The latency and amplitude of P300 was measured (25).

**Assessment of stress parameters:**

Saliva was collected on the recording day, at the beginning of tests and stored at –70°C. Cortisol levels were estimated by ELISA kits.

The mental and emotional acute stress was assessed using Spielberger state and trait anxiety inventory (STAI), a self reported assessment device, administered before the testing for cognitive parameters. The maximum and minimum achievable scores of the STAI are 20 and 80 respectively (26).

The Lead II of ECG was recorded continuously for measuring the changes in heart rate. The galvanic skin response (GSR) or conductivity of the skin was recorded with RMS POLYRITE-D systems. Two Ag/AgCl electrodes around the index and middle finger of the participant's left hand were tied to record the GSR, which is a relatively reliable index of sweat-gland activity and changes in the activation level of the sympathetic nervous system.

60-300 seconds of artifact free data of HR and GSR were used to calculate the mean value, for purpose of comparison between basal and different cognitive tasks. The results of baseline GSR and HR were recorded at the start of the testing procedures on each testing day. ΔGSR and ΔHR were measured as the difference between maximum recorded GSR or HR during performance of various tasks and the baseline GSR or HR recorded during that day.

**Statistical analysis**

Statistical analysis was made using Graphpad Prism 5. All the variables were found to be normally distributed as assessed by D' Agostino and Pearson omnibus normality test. Intra-group comparison was made by Repeated measures ANOVA with Bonferroni post-hoc multiple comparison test and inter-group comparison was made with two tailed, unpaired t-test. $P<0.05$ was considered significant. After the analysis, the blinding was opened and it was revealed that Group A and B were placebo and EtOS respectively.

# Results

**Demographic data and compliance:**

A total of 44 subjects were inducted for the study. The disposition of subjects is represented in Table I. Compliance in taking the capsules each day was greater than 90%. Two subjects in group A and one in group B were lost to follow-up due to work- time restriction and one subject in group A discontinued the medication complaining of nausea. The subjects

TABLE I : Comparison of physical characteristics at baseline.

|  | Placebo (n=20) | EtOS (n=20) | P value |
|---|---|---|---|
| Age (years) | 27.53±2.64 | 27±2.82 | NS |
| Height (cm) | 170.26±7.38 | 170.5±3.3 | NS |
| Weight (kg) | 67.33±5.75 | 65.37±3.05 | NS |
| BMI (kg/m$^2$) | 23.24±1.71 | 22.5±1.22 | NS |

Values are expressed as Mean±SEM. Unpaired t test between Placebo and EtOS groups.

in the groups were not significantly different in their educational qualification, age, height, weight or BMI.

**Sternberg memory test:**

The reaction time (RT) and error rate (ER) of the Sternberg memory scan test are presented in Table II and III respectively.

There was an improvement in the RT of the Sternberg, in both groups, over time. Though the initial improvement at D15 was comparable at 4.3% and 4.9%, the improvement was very marginal between D15 and D30 in the Placebo group (1.3%), as compared to EtOS group (4.3%). The inter-group comparison in RT at D30 was significant.

There was a reduction in the ER of the Sternberg, in both groups, over time. The % change in ER, between D0 and D30, in placebo and EtOS groups was 38

TABLE II : Effects of *Et*OS (n=20) and placebo (n=20) on Reaction time (msec) of Sternberg memory scan test and Stroop test.

| Test | Group | Day 0 | Day 15 | Day 30 | P value |
|---|---|---|---|---|---|
| Sternberg test | Placebo | 931.9±21.58 | 891.4±23.28 P<0.01[a] | 879.7±23.47 P<0.001[b] NS[c] | <0.001 |
|  | EtOS | 892.1±18.99 | 847.7±2.36 P<0.001[a] | 810.6±23.83 P<0.001[b] P<0.01[c] | <0.001 |
|  | P value | NS | NS | <0.05 |  |
| Facilitation task | Placebo | 796.6±21.27 | 791.5±16.54 NS[a] | 767.1±16.59 NS[b]NS[c] | NS |
|  | EtOS | 805.5±25.78 | 756.3±26.53 NS[a] | 725.3±21.80 P<0.01[b] NS[c] | <0.01 |
|  | P value | NS | NS | NS |  |
| Neutral task | Placebo | 832.1±13.50 | 809.7±15.22 NS[a] | 800.8±18.74 P<0.05[b] NS[c] | <0.05 |
|  | EtOS | 827.7±27.04 | 794.8±22.05 NS[a] | 734.9±19.71 P<0.001[b] P<0.01[c] | <0.001 |
|  | P value | NS | NS | <0.05 |  |
| Interference task | Placebo | 930.6±17.11 | 903.9±17.00 NS[a] | 894.9±17.02 P<0.05[b] NS[c] | <0.05 |
|  | EtOS | 949.9±21.76 | 895.0±22.69 P<0.01[a] | 839.7±20.75 P<0.001[b] P<0.01[c] | <0.001 |
|  | P value | NS | NS | <0.05 |  |

Values are expressed as Mean±SEM. NS: not statistically significant, RM: Repeated measures ANOVA with Bonferroni post-hoc multiple comparison test for intragroup comparison.Unpaired t test between Placebo and EtOS groups. [a]Comparison of day 0 and day 15. [b]Comparison of day 0 and day 30. [c]Comparison of day 15 and day 30.

TABLE III : Effects of EtOS (n=20) and placebo (n=20) on Error rate (percentage) of Sternberg memory scan test and Stroop test.

| Test | Group | Day 0 | Day 15 | Day 30 | P value |
|---|---|---|---|---|---|
| Sternbergtest | Placebo | 5.30 (4.00–8.30) | 4.00 (3.00–6.80) P<0.05[a] | 3.30 (1.92–5.18) P<0.001[b] P<0.05[c] | <0.001 |
| | EtOS | 4.68 (4.05–8.30) P<0.05[a] | 3.67 (2.00–6.10) | 1.60 (0.00–3.80) P<0.001[b] P<0.01[c] | < 0.001 |
| | P value | NS | NS | <0.05 | |
| Facilitation task | Placebo | 1.00 (0.00–2.75) | 1.30 (0.00–1.39) NS[a] | 0.00 (0.00-1.00) NS[b] NS[c] | <0.05 |
| | EtOS | 0.05 (0.00–2.68) | 1.30 (0.00–1.39) NS[a] | 0.00 (0.00-1.00) NS[b] NS[c] | <0.01 |
| | P value | NS | NS | NS | |
| Interference task | Placebo | 2.89 (1.39–5.5) | 2.08 (1.40–4.17) NS[a] NS[a] | 1.89 (1.39-2.78) NS[b] NS[c] | <0.05 |
| | EtOS | 4.05 (1.25–5.89) | 2.78 (0.25–4.17) NS[a] | 1.34 (0.10-2.78) <0.001[b] NS[c] | <0.001 |
| | P value | NS | NS | <0.05 | |

Values are expressed as Median (Interquartile range). NS: not statistically significantFriedman test with Dunn's multiple comparison for intragroup comparison.Mann-Whitney U test between Placebo and EtOS groups. [a]Comparison of day 0 and day 15. [b]Comparison of day 0 and day 30. [c]Comparison of day 15 and day 30.

and 66 respectively. The inter-group comparison in ER at D30 was significant

**Stroop test:**

The RT and ER of Stroop task are tabulated in Tables II and III respectively.

**Facilitation task:**

Though there was an improvement in the RT of Facilitation task in the EtOS group only, ER reduced in both the placebo and EtOS groups. The inter-group comparisons of RT and ER at D15 or D30 were not significant.

**Neutral task:**

There was an improvement in the RT of Neutral task in both the placebo and EtOS groups over time. At D30, the percentage change in RT, as compared to D0, in the placebo and EtOS groups was 3.7 and 11.2 respectively. Interestingly, as in the Sternberg

test, the improvement was minimal between D15 and D30 in the placebo group (1.1%) as compared to the EtOS group (7.5%). The inter-group comparison in RT at D30 was significant.

The inter and intra-group comparisons of ER of Neutral task at D15 or D30 were not significant (data not given).

**Interference task:**

There was an improvement in the RT of interference task, in both the Placebo and EtOS groups. The percentage change in RT between D0 and D30 were 3.8 and 11.6 in the placebo and EtOS groups respectively. Similar to the Sternberg test and neutral task of Stroop test, there was only a minimal improvement between D15 and D30 in the placebo group (0.9%) as compared to the EtOS group (6.1%). The inter-group comparison in RT at D30 was significant.

Though there was a significant reduction in the ER

of interference task in both the groups, the percentage change in ER, between D0 and D30, were 35 and 66 in the placebo and EtOS groups respectively. The inter-group comparison in ER at D30 was significant.

**Event related potential:**

P300 latency was comparable at D0 in the placebo (286.7±18.23 ms) and EtOS groups (284.5±21.86 ms). There was a significant reduction of the P300 latency in the EtOS group alone to 270.9±21.78 ms on D30 (p<0.01). This comparison was significant between D15-30 (p<0.05) and D0-30 (p<0.001). The inter-group comparison of P300 at D30 was significant (p=<0.05).

The P300 amplitude was not significantly different in two groups at any point of time (Data not included).

**Salivary cortisol:**

The baseline salivary cortisol levels were comparable at 7.15±3.27 and 7.16±3.60 nmol/L in the placebo and EtOS groups respectively. There was a significant (p<0.001) reduction in the salivary cortisol values in the EtOS group alone (14%) This reduction was significant only after D15. The inter-group comparisons at D15 or D30 were not significant.

**State and Trait Anxiety Inventory (STAI):**

The scores of State component of STAI were comparable at D0 in the placebo (32.70±7.93) and EtOS groups (34.30±7.01), (p>0.05). There was a significant reduction in the scores in the EtOS group alone to 29.45±6.17 (p<0.001). This comparison was significant only between D0-D30 (p<0.001). The inter-group comparison was not significant at D15 or D30.

The scores of Trait component of STAI were comparable in the placebo (34.85±4.17) and EtOS groups (36.80±5.54) (p=NS). There was a significant reduction in the scores in the EtOS group alone to 32.95±4.51 (p<0.001). This comparison was significant between D15-30 (p<0.05) and D0-30 (p<0.001).

**GSR and HR:**

The baseline GSR and HR at D0 in the placebo and EtOS groups were comparable at 9.30±1.30 µS, 69.70±10.15 per min and 9.11±1.14 µS, 70.65±8.71 per min respectively. The ΔGSR and ΔHR at D0 in the placebo and EtOS groups were 0.75±0.52 µS, 29.55±8.51 per min and 0.79±0.55 µS, 27.75±10.83 per min respectively. There were no significant inter-group or intra-group differences in any of these parameters on D15 or D30.

## Discussion

The effects of EtOS on stress and cognition were studied on 40 healthy human volunteers by a double-blinded randomized control study over 30 days.

Results of the study showed that both placebo and EtOS intake over a period of 30 days resulted in a significant improvement in the RT of Sternberg test, neutral and interference task of Stroop test. The ER of Sternberg test, facilitation and interference task of Stroop test also showed improvement in both the groups. 'Practice effect' is a well known phenomenon wherein familiarization and repeated testing of the same cognitive parameter in a sub-group increased responsiveness and performance (27, 28). It is worth noting that the improvement in the placebo group tended to plateau after D15, while in the EtOS group there was a continued significant improvement beyond the D15. Also, inter-group comparison of RT and ER of Sternberg and Interference task, and RT of Neutral task in the EtOS group, showed that EtOS had a beneficial effect over and above the Placebo. Sternberg test, a parameter to assess short term memory and central executive functions of cognition, represents information processing and retrieval of information (23). Stroop task is a parameter of cognitive flexibility, represents goal-directed working memory (24). The extract of *Ocimum sanctum* seems to have the potential to improve short term memory and cognitive flexibility. This is the first study to document the enhancement of cognitive performance of the subjects after intake of OS for a short period.

In animal models of Alzheimer's disease, extract of OS has been shown to improve learning and memory deficits, improve spatial learning and alleviate the associated neuropsychological symptoms. The beneficial effect observed with OS have been attributed to its anti-oxidant activity in animal studies (5). The active principle of the ethanolic leaf extract of OS used in this study contains 2.7% ursolic acid, a compound, which has antioxidant properties and gives remarkable protection against lipid peroxidation (29, 30).

The P300 latency, an indicator of memory capacity and attention (31), remained fairly constant in the placebo group. However the significant improvement in the EtOS group, beyond D15, indicates that EtOS had a favorable effect on attention.

In the placebo group, though there was an improvement of the RT, P300 latency remained fairly constant. However EtOS showed improvement in both RT and P300. P300 reflects relative timing of stimulus evaluation whereas RT reflects relative timing of response processes (32). The differential response of OS to RT and P300 could provide a lead point to probe into the key neuronal processes through which OS may act.

Our study group predominantly contained academically successful young adults, who had good cognitive capacity. It is interesting to note that administration of EtOS could improve the cognitive parameters of those even in this population.

Salivary cortisol levels significantly decreased in the EtOS group alone. But the lack of inter-group differences and the magnitude of change, indicate that this may not be clinically significant. Cortisol is a hormone whose concentrations are extremely variable and subjected to important circadian variation. One single measurement in a day is not significant, as it could be altered by an extremely high number of variables. Measurement of cortisol awakening response or repeated measurements of salivary cortisol levels could have thrown better light and we consider it a limitation of the study with respect to this parameter.

The scores of state and trait components of STAI were significantly decreased in the EtOS group alone, but there were no differences on inter-group comparison. The healthy young individuals included in the study seemed to have low-normal baseline score of STAI indicating there did not suffer from a lot of anxiety. Inclusion of aged volunteers who may suffer from stress may be useful to understand the real time significance. There were no significant differences in basal or ΔHR and GSR responses between the groups. Thus the effects *Ocimum sanctum* of on stress could not be established in the population with the current dose and parameters used.

A reduction in stress in 71 subjects having symptoms of stress (21) and reduction in anxiety and depression in patients of generalized anxiety disorder (16) has been documented earlier based on self reported questionnaires with a higher dose for longer period. Estimation of cortisol was not done in these studies though the authors stated that major action of *Ocimum Sanctum* in stress related disorder is via modulation of hypothalamus-pituitary-adrenocortical axis. Thus, there are no objective studies available to assess the cognitive enhancement and anti- stress effect of OS in humans.

Thus, *Ocimum sanctum* does seem to have a favorable effect on certain cognitive parameters like short term memory, cognitive flexibility and attention. Clinical significance of these effects could be verified by conducting the study with a larger population in vulnerable age group and by increasing the dose and duration of intake of *Ocimum Sanctum*.

**Conflict of Interest:**

No conflict to disclose

# Acknowledgements

# References

1. Singh N, Verma P, Pandey BR, Bhalla M. Therapeutic potential of *Ocimum sanctum* in prevention and treatment of cancer and exposure to radiation: An Overview. *International Journal of Pharmaceutical Sciences & Drug Research* 2012; 4(2): 97–104.

2. Sakina MR, Dandiya PC, Hamdard ME, Hameed A. Preliminary psychopharmacological evaluation of *Ocimum sanctum* leaf extract. *Journal of Ethnopharmacology* 1990; 28(2): 143–150.

3. Joshi H, Parle M. Evaluation of nootropic potential of *Ocimum sanctum* Linn. in mice. *Indian Journal of Experimental Biology* 2006; 44(2): 133–136.

4. Giridharan, Vayalanellore V, Thandavarayan RA et al. *Ocimum sanctum* Linn. Leaf extracts inhibit acetylcholinesterase and improve cognition in rats with experimentally induced dementia. *Journal of Medicinal Food* 2011; 14(9): 912–919.

5. Raghavendra M, Maiti R, Kumar S, Acharya SB. Role of *Ocimum sanctum* in the experimental model of Alzheimer's disease in rats. *International Journal of Green Pharmacy* 2009; 3(1): 6. doi:10.4103/0973-8258.49368.

6. Yanpallewar SU, Rai S, Kumar M, Acharya SB. Evaluation of antioxidant and neuroprotective effect of *Ocimum sanctum* on transient cerebral ischemia and long-term cerebral hypoperfusion. *Pharmacology, Biochemistry, and Behavior* 2004; 79(1): 155–164.

7. Ahmad A, Khan MM, Raza SS et al. *Ocimum sanctum* attenuates oxidative damage and neurological deficits following focal cerebral ischemia/reperfusion injury in rats. *Neurological Sciences* 2012; 33: 1239–1247.

8. Sembulingam K, Sembulingam P, Namasivayam A. Effect of *Ocimum sanctum* Linn on noise induced changes in plasma corticosterone level. *Indian J of Physiol Pharmacol* 1997; 41(2): 139–143.

9. Archana R, Namasivayam A. Effect of *Ocimum sanctum* on noise induced changes in neutrophil functions. *Journal of Ethnopharmacology* 2000; 73 (1-2): 81–85.

10. Maity TK, Mandal SC, Saha BP, Pal M. Effect of *Ocimum sanctum* roots extract on swimming performance in mice. *Phytotherapy Research* 2000; 14(2): 120–121.

11. Muruganandam AV, Kumar V, Bhattacharya SK. Effect of poly herbal formulation, EuMil, on chronic stress-induced homeostatic perturbations in rats. *Indian Journal of Experimental Biology* 2002; 40(10): 115–160.

12. Tabassum, Imrana, Siddiqui ZN, Rizvi SJ. Effects of *Ocimum sanctum* and camellia sinensis on stress-induced anxiety and depression in male albino rattus norvegicus. *Indian Journal of Pharmacology* 2010; 42(5): 283–288.

13. Chatterjee M, Verma P, Maurya R, Palit G. Evaluation of ethanol leaf extract of *Ocimum sanctum* in experimental models of anxiety and depression. *Pharmaceutical Biology* 2011; 49(5): 477–483.

14. Gupta P, Yadav DK, Siripurapu KB et al. Constituents of *Ocimum sanctum* with antistress activity. *Journal of Natural Products* 2007; 70(9): 1410–1416.

15. Samson J, Sheeladevi R, Ravindran R. Oxidative stress in brain and antioxidant activity of *Ocimum sanctum* in noise exposure. *Neurotoxicology* 2007; 28(3): 679–685.

16. Bhattacharyya D, Sur TK, Jana U, Debnath PK. Controlled programmed trial of *Ocimum sanctum* leaf on generalized anxiety disorders. *Nepal Medical College Journal* 2008; 10(3): 176–179.

17. Ravindran R, Rathinasamy SD, Samson J, Senthilvelan M. Noise-stress-induced brain neurotransmitter changes and the effect of *Ocimum sanctum* (Linn) treatment in albino rats. *Journal of Pharmacological Sciences* 2005; 98(4): 354–360.

18. Samson J, Rathinasamy SD, Ravindran R, Senthilvelan M. Biogenic amine changes in brain regions and attenuating action of *Ocimum sanctum* in noise exposure. *Pharmacology, Biochemistry, and Behavior* 2006; 83(1): 67–75.

19. Sembulingam K, Sembulingam P, Namasivayam A. Effect of *Ocimum sanctum* Linn on the changes in central cholinergic system induced by acute noise stress. *Journal of Ethnopharmacology* 2005; 96(3): 477–482.

20. Pandey R, Gupta S, Shukla V, Tandon S. Antiaging, antistress and ROS scavenging activity of crude extract of *Ocimum sanctum* (L.) in caenorhabditis elegans. *Indian Journal of Experimental Biology* 2013; 51(7): 515–521.

21. Saxena RC, Singh R, Kumar P et al. Efficacy of an extract of Ocimum Tenuiflorum (Oci Best) in the management of general stress: A Double-Blind, Placebo-Controlled Study. *Evidence-Based Complementary and Alternative Medicine*: 2012; 894509.

22. Mondal S, Varma S, Bamola VD et al. Double-blinded randomized controlled trial for immunomodulatory effects of tulsi (*Ocimum sanctum* Linn.) Leaf extract on healthy volunteers. *Journal of Ethnopharmacology* 2011; 136(3): 452–456.

23. Sternberg S. High-Speed Scanning in Human Memory. *Science* 1966; 153(3736): 652–654.

24. MacLeod C M. Half a century of research on the stroop effect: An Integrative Review. *Psychological Bulletin* 1991; 109(2): 163–203.

25. Patel SH, Azzam PN. Characterization of N200 and P300: selected studies of the event-related potential. *International Journal of Medical Sciences* 2005; 2(4): 147–154.

26. Spielberger CD. Manual for the StateTrait Anxiety Inventory (Form Y). Palo Alto, CA: Mind Garden 1983.

27. Newell A, Rosenbloom PS. Mechanisms of Skill acquisition and the law of practice. Cognitive skills and their acquisition 1, 1980.

28. Delaney PF, Reder LM, Staszewski JJ, Ritter FE. The Strategy-specific nature of improvement: The power law applies by strategy within task. *Psychological Science* 1998; 9(1): 17.

29. Liu J. Pharmacology of oleanolic and ursolic acid. *J Ethnopharmacol* 1995; 49: 57–68.

30. Pemminati S, Nair V, Dorababu P, Gopalakrishna H N, Pai M. Effect of ethanolic leaf extract of Ocimum sanctum on haloperidol-induced catalepsy in albino mice. *Indian J Pharmacol* 2007; 9: 87–89.

31. Yamaguchi S, Knight RT. P300 Generation by Novel Somatosensory Stimuli. *Electroencephalography and Clinical Neurophysiology* 1991; 78 (1): 50–55.

32. Kutas M, McCarthy G, Donchin E. Augmenting mental chronometry: The P300 as a measure of stimulus evaluation time. *Science* 1977; 197(4305): 792–795.