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Aromatherapies and Stress: An Examination of Saliva Sclarea in Acute Stress Conditions

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AROMATHERAPIES AND STRESS: AN EXAMINATION OF SALVIA SCLAREA IN ACUTE STRESS CONDITIONS

A Master's Thesis

Presented to

The Graduate College of

Missouri State University

In Partial Fulfillment

Of the Requirements for the Degree

Master of Science, Psychology

By

Emily J. Van Gundy

May 2024

AROMATHERAPIES AND STRESS: AN EXAMINATION OF SALVIA SCLAREA IN ACUTE STRESS CONDITIONS

Psychology

Missouri State University, May 2024

Master of Science

Emily J. Van Gundy

ABSTRACT

The use of essential oils as an alternative medicine in the United States is becoming increasingly popular as more accreditation is given to its therapeutic values. With this increase in use, we must continue to examine the uses of essential oils, including associated risks. One unavoidable aspect of life is stress, both acute and chronic. This study aims to better our understanding of the stress-reducing properties of the essential oil clary sage (salvia sclarea) after an experimentally induced acute stress experience. The Trier Social Stress Test was used to elicit an acute stress response. Participants were given the scent of either clary sage essential oil or sweet almond oil post-stressor. Psychological and physiological measures of stress were measured. Paired samples independent t-tests (sweet almond oil control group; clary sage experimental group) were used to examine differences in salivary cortisol, blood pressure, and responses to the State Trait Anxiety Inventory. Current results indicate that clary sage does not affect the reduction of salivary cortisol presence. No significant difference in blood pressure has been observed at this time, though results are trending towards the clary sage group having a significant decrease. No difference in responses to the STAI was found following the administration of either clary sage essential oil or sweet almond oil. Further research is needed to understand the potential anxiolytic effects of clary sage, and its use as a secondary treatment for anxiogenic disorders.

KEYWORDS: aromatherapy, essential oils, acute stress, perception, olfaction

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In the interest of academic freedom and the principle of free speech, approval of this thesis indicates the format is acceptable and meets the academic criteria for the discipline as determined by the faculty that constitute the thesis committee. The content and views expressed in this thesis are those of the student-scholar and are not endorsed by Missouri State University, its Graduate College, or its employees.

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Introduction

Aromatherapy techniques are seen in many societies and cultures and have been used for centuries. Currently in the United States, aromatherapy, as seen in the use of essential oils, is viewed as an alternative medicine. Essential oils such as lavender (lavandula angustifolia), lang-ylang (Cananga odorata), and jasmine (jasminum grandiflorum) are reported to promote relaxation. Other essential oils are said to improve memory, concentration, and affect (Farrar & Farrar, 2020).

When essential oil molecules are inhaled, molecules of the substance travel through the nostrils to the olfactory bulb. From here the brain further processes the stimulus in the amygdala and hippocampus. The amygdala often triggers an emotional response to the stimulus, and the hippocampus associates the stimulus with memories. The limbic system interacts with the cerebral cortex to give an individual a conscious experience of the stimuli. Scents may also exert direct effects on neuronal receptors by crossing the blood-brain barrier. Within the respiratory system, when essential oil molecules are inhaled, they travel to the upper and lower respiratory tract. From here the inhaled molecules enter the bloodstream (Farrar & Farrar, 2020; Hartley & McLachlan, 2022).

Essential Oil Anxiolytic Effects

Among essential oils, previous research has indicated that lavender and chamomile (matricaria recutita) essential oils are effective in reducing the severity of symptoms in psychiatric disorders, such as depression and anxiety, as well as reducing feelings of stress (Tsang et al., 2015; Fung & Tsang, 2017). Fung and Tsang (2017) pose that aroma-massage (i.e. massage practices utilizing scented essential oils) may be beneficial for reducing distress seen in dementia patients. Here authors utilized a three-arm parallel group design. Three groups were

examined, each experiencing different treatments for behavioral and psychological symptoms associated with dementia. Group 1 received aroma-massage with acupressure, group 2 received cognitive training and exercise, and group 3 received aroma-massage with acupressure and cognitive training. This study defines exercise as "treatment as usual" for all groups. Results found by Fung and Tsang (2017) state that groups 1 and 3, those containing aroma-therapy massage with acupressure, had a significant reduction in affective and physical distress associated with dementia. Group 2 was not reported to share this reduction. The use of essential oil aromatherapy has also been investigated outside of special populations, such as that of individuals with dementia.

Tsang and colleagues (2015) examined a multicomponent stress management program involving cognitive behavioral therapy techniques, relaxation exercises, aromatherapies, and acupressure in elementary school teachers with symptoms of depression or anxiety in their pilot study. The intervention group had a significant reduction in depression, anxiety, and stress when compared with the wait-list control group. This quasi-experimental study did not examine the effects of aromatherapy alone. Further research is needed to investigate the effects of individual treatments.

In a meta-analysis consisting of 21 articles, lavender essential oil was examined for significant anxiolytic, or anxiety reducing, properties. There were 791 participants in the control group, and 804 in the experimental group. It was found that lavender essential oil treatment significantly decreased participants' stress score ratings in the experimental group when compared to the control group (Ghavami et al., 2022). Other essential oil scents have also been investigated for anxiolytic properties, such as (rosmarinus officinalis L.), jasmine, and fir (abies alba).

Rosemary essential oil inhalation is suggested to reduce feelings of stress or anxiety. After being exposed to the scent of rosemary essential oil, rats were then subjected to a tail suspension test which acted as an acute stressor. It was found that rats who previously smelled rosemary essential oil had less serum corticosterone and higher dopamine levels following acute stress exposure. This study suggests that these results are due to the anxiolytic properties of rosemary, specifically α-pinene (Villareal et al., 2017). Physiological responses to essential oil aromatherapy have also been examined within the scope of galvanic skin conductance, skin temperature, and respiration rate.

Kuo (2017) examined the impact of the scent of jasmine on the central nervous system. Participants were directed to inhale the scent of jasmine for five minutes. It was found that after exposure to jasmine, participants' respiration rate and peripheral skin temperature decreased, and skin conductance measures increased. Pain threshold was also assessed, and it was found that after inhaling the scent of jasmine participants had a higher pain tolerance (Kuo, 2017). From this study, it is suggested that jasmine essential oil inhalation may promote feelings of relaxation in users, as well as help regulate the central nervous system. Here the physiological responses of essential oil aromatherapy were examined. Psychological responses to essential oil scents, that of fir, have also been investigated.

Kim and Song (2022) discuss the impact of fir essential oil scent on feelings of negative affect. It was found that following administering the scent of fir essential oil for three minutes, participants reported significantly less state and trait anxiety on the State Trait Anxiety Inventory (STAI), compared to the control group (Kim & Song, 2022). The results of this study further suggest the usefulness of essential oil scent inhalation in reducing negative affectivity.

Given the research regarding the anxiolytic benefits of essential oils, aromatherapy may have the capability of providing the user with a feeling of relaxation and reducing measurements of biological stress (Kim & Song, 2022; Kuo, 2017; Villareal et al., 2017; Ghavami et al., 2022). Little research has been done regarding the therapeutic value of clary sage essential oil scent. The current study aims to examine clary sage's impact on decreasing self-report feelings and physiological measures of anxiety.

Salvia Sclarea Anxiolytic Effects

Clary sage is one of many plants within the genus Salvia. Salvia plants are rich in polyphenol compounds, such as flavonoids and phenolic acids. These plants also contain essential oils with a variety of terpenoids, such as α and β-Thujone, camphor 1, 8-cineole, α-humulene, and β-caryophyllene among others. Plants within the genus Salvia also commonly contain polysaccharides such as arabinogalactans and pectin. Plants within the genus Salvia have neurotrophic, anti-inflammatory, anti-depressant, anxiolytic, and antioxidant effects due to the components' neurological and biochemical effects (Lopresti, 2017). Very little research has been directed toward investigating the effects of clary sage on acute and chronic stress states in human participants.

Gross and colleagues (2013) investigated the chronic administration of clary sage essential oil in mice. Mice were grouped based on dominant or submissive behavior, and if they received clary sage essential oil or sunflower oil. Authors found that dominant mice had a reduction in dominant and anxiety-like behaviors when given the clary sage essential oil in their food, compared to the dominant mice given sunflower oil. Submissive mice that were given clary sage essential oil also displayed a decrease in anxiety-like behaviors and reduced corticosterone (Gross et al., 2013). It is suggested that this reaction is due to the anxiolytic effects of clary sage.

Seol et al. (2010) also examined the effects of clary sage in rats following an acute stressor task. Here, rats were presented with several scents, clary sage included, through either inhalation or injection, then were made to participate in a forced swim task (FST). The stressor here was assessed by timing how long it took rats to begin swimming in the FST. It was found that the clary sage serum had the strongest anti-stressor effects among the essential oils tested (Seol et al., 2010). Rats in the clary sage group had the shortest duration of immobility time when beginning the FST. This further suggests the therapeutic benefits of aromatherapy, specifically in the use of clary sage for stress or anxiety reduction. Authors pose that these effects of clary sage were mediated via dopaminergic pathways due to their involvement in movement. In addition to the anxiolytic effects of clary sage on animal participants, research with human participants has also supported the effects of clary sage.

One double-blind study by Seol et al. (2013) examined a group of female participants undergoing urodynamic examinations, an acute stress experience. There was one control group and two experimental groups. Within the experimental group participants either inhaled the scent of lavender or the scent of clary sage. The control group was given the scent of sweet almond oil. This study concluded that participants who inhaled the scent of clary sage had a significant decrease in systolic and diastolic blood pressure compared to the control group. A significant reduction in respiratory rate was also observed in the clary sage group (Seol et al., 2013). The results of this study suggested that the scent of clary sage may promote feelings of relaxation and stress reduction in those undergoing stressful medical examinations. Clary sage has also been examined for potential hypertension reducing properties outside of acute stress conditions. Research has suggested that clay sage inhalation may reduce heart rate.

A study conducted by Mitic et al (2020) found that after participants inhaled the scent of clary sage for 30 minutes, females had a significant reduction in post-test heart rate. The authors also found that male participants had a significant reduction in heart rate, but less so than females (Mitic et al., 2020). Current research regarding the potential anxiolytic effects of clary sage essential oil aromatherapy is minimal, both within animal and human studies.

The Current Study

Many essential oil compounds have been shown to have psychological and physiological impacts on both human and animal participants (Tsang et al., 2015; Fung & Tsang, 2017; Ghavami et al., 2022, & Gross et al., 2013). Due to the minimal literature that has examined clary sage, as well as its reported ability to have central nervous system calming properties and anxiolytic effects, this study examines the effects of clary sage essential oil scent under acute stress conditions, utilizing the Trier Social Stress Test (TSST) (Kirschbaum et al., 1993). The TSST is commonly used to elicit an acute stress response from participants in laboratory conditions. The TSST relies heavily on social evaluative threat and uncontrollability (Dickerson & Kemeny, 2004).

The current study hypothesizes that participants in the clary sage experimental group will have a greater reduction in blood pressure at assessment time points compared to the sweet almond oil control group (Seol et al., 2013). Furthermore, it is predicted that salivary cortisol measures in the clary sage experimental group will have a greater reduction in cortisol presence compared to the sweet almond control group, indicating that the scent of clary sage is exhibiting anxiolytic effects (Seol et al,. 2013; Nuiden et al., 2021).

Within affective assessment, it is predicted that participants' responses to the short form of the State Trait Anxiety Inventory (STAI-6) (Tluczek et al., 2009) will indicate significantly

less state anxiety within the clary sage experimental group compared to the sweet almond oil control group.

Method

Participants

All participants in this study came from an introductory psychology class at Missouri State University. There was a total of 60 participants, the majority of which identified as female (61.9%) and White (68.9%). Participants' average age was 20.4 years with a standard deviation of 5.24. Data was analyzed on 38 participants' salivary cortisol samples, and 51 participants' blood pressure measures.

An a priori power analysis was conducted using G*Power (Faul et al., 2007) to determine the minimum sample size. Results indicated that at 80% power, at a medium effect size, ($\alpha = .05$), 102 participants are needed for a t-test of independent means.

Materials

Participants were asked to complete a demographics survey which included questions on age, ethnicity, educational level, and gender (Appendix A).

The STAI-6 was used post-test and at times of saliva collection to assess participants' subjective feelings of state anxiety (Tluczek et al., 2009). The STAI-6 consists of 6 questions with four-point Likert scale response options (1 = not at all; 4 = very much). STAI-6 contains questions such as "I feel calm" and "I am tense". This questionnaire has a Cronbach's alpha greater than .90 (Tluczek et al., 2009).

The DASS21 (Lovibond & Lovibond, 1995) was used to screen for indicators of depression which this study will control for, as well as to further examine feelings of stress and anxiety in participants within the past seven days. This questionnaire consists of 21 statements

such as “I found it difficult to work up the initiative to do things” and “I felt that I had nothing to look forward to”. Participants may respond to these statements using a 4-point Likert scale (0 = did not apply to me at all; 3 = applied to me very much or most of the time). The DASS21 has been shown to have a Cronbach’s alpha of 0.76 to 0.90 (Thi Hong Lee et al., 2017).

The TSST was used to elicit an acute stress response as an experimental condition. The TSST consists of a three-minute anticipatory period, a five-minute speech task, and a five-minute mental arithmetic task (Kudielka et al., 2007; Kirschbaum et al., 1993). Participants were asked to perform a mock job interview and a mental arithmetic task in front of a panel of judges. There was one in-person judge. A video recording of two individuals acting to be judges remotely observing the experiment was played during the TSST. The tasks within the TSST have elements of uncontrollability and social evaluative threat, which elicit a greater cortisol response than speech and mental arithmetic tasks without these elements (Dickerson & Kemeny, 2004). Due to participants being told there are two additional remote judges, aside from the one in-person judge, participants experienced a higher degree of social evaluative threat. Aside from an increase in social evaluative threat, participants performed tasks to both male and female presenting judges. This allowed both male and female participants to experience social evaluative threats from a judge of their own sex.

Activation of the HPA axis in acute stress conditions was assessed by analyzing participants’ salivary cortisol levels (Hellhammer et al., 2009). This was assessed under the Salimetrics expanded range high sensitivity salivary cortisol enzyme immunoassay protocol (Salivary cortisol elisa kit, 2024). Cortisol is a glucocorticoid that is produced by the adrenal cortex. Saliva samples were collected using the self-collection passive drool method. This method requires the participant to salivate through a straw into a vial (Fernandes et al., 2013).

Participants were required to collect at least 1.8 milliliters of saliva. Following collection, samples were to be stored at -20ºC. Due to the diurnal rhythm of cortisol, saliva samples from participants will only be gathered from 12PM through 6PM. Salivary cortisol concentrations gradually increase in the morning, peaking 30 minutes after an individual wakes up. Concentration then decreases throughout the day (Kobayashi et al., 2017). Participants were questioned if they had consumed any food or drink for before participating. This includes alcoholic beverages in the past 12 hours, and those that contain caffeine within the past one hour. Participants were also questioned as to if they had smoked within the past hour, if they had a recent infection, and if they had experienced any social problems within the past three hours.

Participants had their systolic and diastolic blood pressure measured using the automatic oscillometric technique, which is commonly viewed as a gold standard in measuring human blood pressure (Chen et al., 2017). Participants had a blood pressure cuff attached to their left upper arm. Once the "start" button was pressed, the cuff began automatically inflating. Once the device determined a blood pressure reading, it automatically deflated. Blood pressure was only measured at assessment time points. The researcher did not manually determine blood pressure.

The present study used a small cotton pad with one milliliter of Handcraft Blends 100% pure clary sage essential oil or US+ pure 100% sweet almond oil. Participants were instructed to hold this at their nose for five minutes. Commonly sweet almond oil is used as a base or dilutant in the use of other essential oils. Previous research has used sweet almond oil for a detectable control when examining aromatherapies (Seol et al., 2013). Furthermore, no physiological effects of sweet almond oil have been found (Nuiden et al., 2021).

Procedure

All research conducted in this paper was conducted following APA's guidelines for the ethical treatment of human participants and was reviewed and approved by the Institutional Review Board (IRB) of Missouri State University (Appendix B).

Participants in this study were asked to sign an informed consent document. Participants were allowed to ask any questions before beginning study participation. Participants then provided a baseline salivary cortisol sample and had their blood pressure measured. Participants then completed the STAI. This took approximately 10 minutes.

Immediately following this, participants began the TSST. This consisted of a three-minute anticipatory period, a five-minute speech task, and a five-minute arithmetic task (Kudielka et al., 2007). In the anticipatory period, participants were asked to prepare for the speech task. The speech task was a mock job interview. Participants were instructed to speak as if they were interviewing for a desired job, and to continuously speak for five minutes. In the arithmetic task participants were asked to subtract 13 from 1037 repeatedly (1037 – 13 = 1024, 1024 – 13) for five minutes. If a participant responded incorrectly to this arithmetic task, they were asked to start over from 1037. A video recording of two individuals acting to be judges in the TSST was presented to participants, with one in-person judge present. Participants were shown the virtual judges at the start of the TSST briefly. The computer screen was then turned away from the participants' view and a webcam was positioned to illude to participants that they were being viewed by the judges. Participants were not recorded. During the TSST judges maintained a neutral disposition and only gave feedback during the mental arithmetic task if a response was incorrect. This task took approximately 15 minutes. Following completion of the TSST participants had their blood pressure measured and completed the STAI-6. Once this was

completed, participants were asked to give a second saliva sample to measure their peak cortisol response. This took approximately 20 minutes.

Stratified randomization was used to divide participants into either the sweet almond oil control or the clary sage experimental group. Participants were not told what group they were in, or what scent they would be smelling. Participants were told that they may or may not detect a scent on the cotton pad, but to inhale from the cotton pad for five minutes. Participants were given clary sage essential oil if in the experimental condition, and sweet almond oil if in the control group.

Following the five-minute inhalation period, participants had their blood pressure measured and then were asked to complete the STAI-6. Once the 20-minute time period after the start of inhalation was reached participants were asked to give a third saliva sample, which assessed the onset of the decrease in cortisol presence. Participants were given nature magazines to read during periods of waiting to act as a neutral stimulus. Once this was collected participants had their blood pressure measured. Participants then completed the STAI-6 and demographics survey. Participants then had 20 minutes to complete a blood pressure measurement and survey assessments. A final saliva sample was then collected to observe a return to baseline. Participants were then asked to complete the DASS21 and debriefed.

Results

Preliminary analysis was conducted using paired samples t-tests in IBM SPSS Statistics for Windows (IBM Corp., 2020). Mahalanobis distances were calculated to examine the data for outliers. No outliers were found to be present, all data was included in the analysis. No corrections were made as a result of Levene's test for homogeneity. A Shapiro-Wilk test of normality suggested a deviation from normality. A logistic transformation of the STAI-6 data was performed.

Time point 2, immediately following the TSST, timepoint 3, 20 minutes following the TSST, and time point 4, 40 minutes following the TSST, were examined for a hypothesized reduction in salivary cortisol presence in the clary sage essential oil group compared to the sweet almond oil group. From time point 2, immediately following the TSST, to timepoint 3, 20 minutes following the TSST, preliminary analysis revealed no significant difference $t(36) = .547$, $p = .588$ in salivary cortisol presence. From timepoint 2 to time point 4, 40 minutes following the TSST, no significant difference was observed $t(36) = .214$, $p = .832$ (Table 1).

Table 1. Salivary Cortisol Descriptive Statistics

	Cortisol Statistics				
Time Point	Group	*N*	*M*	*SD*	*SE*
Time 4 and 2	S	19	0.043	0.116	0.027
	A	19	0.034	0.132	0.03
Time 4 and 3	S	19	0.023	0.098	0.022
	A	19	0.033	0.09	0.021
Time 3 and 2	S	19	0.02	0.108	0.025
	A	19	0.002	0.101	0.023
Time 2	S	19	0.714	0.215	0.049
	A	19	0.62	0.168	0.038
Time 3	S	19	0.734	0.259	0.059
	A	19	0.622	0.186	0.043
Time 4	S	19	0.757	0.217	0.05
	A	19	0.654	0.19	0.044

Blood pressure was hypothesized to decrease in the clary sage group at time points 2, 3, and 4. Preliminary data analysis has shown no significant difference between the clary sage and sweet almond oil group at time point 2 and 3 in systolic $t(49) = -.656$, $p = .515$ and diastolic blood pressure $t(49) = -1.255$, $p = .215$. No significant difference was observed between groups at time point 2 and 4 in systolic $t(49) = -1.144$, $p = .258$ and diastolic blood pressure $t(49) = -1.048$, $p = .30$ (Table 2).

Table 2. Blood Pressure Descriptive Statistics

	Blood Pressure Statistics				
Time Point	Group	*N*	*M*	*SD*	*SE*
Diastolic BP Time 3 and 2	S	25	-1.76	7.361	1.472
	A	26	3.5	19.67	3.858
Diastolic BP Time 4 and 2	S	25	-3.96	6.611	1.322
	A	26	-1.5	9.787	1.919
Systolic BP Time 3 and 2	S	25	-6.48	7.456	1.491
	A	26	-3.692	19.927	3.908
Systolic BP Time 4 and 2	S	25	-6.48	8.963	1.793
	A	26	-3.577	9.157	1.796

Analysis of the STAI-6 data revealed no significant differences following scent administration in the clary sage or sweet almond oil groups (Table 3 and Table 4). A Shapiro-Wilk test of normality suggested a deviation from normality. Logistic transformation of the STAI-6 scale data was performed.

Table 3. STAI-6 Descriptive Statistic Results

	STAI-6 Descriptive Statistics				
Time Point	Group	*N*	*M*	*SD*	*SE*
Time 2	S	25	13.8	1.354	0.271
	A	26	13.538	1.964	0.385
Time 3	S	25	13.88	1.236	0.247
	A	26	13.577	1.793	0.352
Time 3 and 2	S	25	0.08	0.862	0.172

Table 4. STAI-6 T-Test Results

	STAI-6 T-Tests		
Time Point	*t*	*df*	*p*
Time 2	0.551	49	0.584
Time 3	0.7	49	0.487
Time 3 and 2	0.146	49	0.885

No significant correlation was observed between depression score, measured through responses on the DASS21, and salivary cortisol, blood pressure, and responses to the STAI-6. (Table 5).

Table 5. Depression Score Correlations

	Depression Score		
Variable	Pearson Correlation	*p*	*N*
Cortisol Time 2	-0.109	0.513	38
Cortisol Time 3	0.502	0.502	38
Cortisol Time 4	-0.107	0.522	38
Systolic BP Time 1	-0.048	0.738	51
Diastolic BP Time 1	-0.26	0.065	51
Systolic BP Time 2	-0.211	0.137	51
Diastolic BP Time 2	-0.074	0.607	51
Systolic BP Time 3	-0.021	0.882	51
Diastolic BP Time 3	-0.048	0.737	51
Systolic BP Time 4	-0.222	0.117	51
Diastolic BP Time 4	-0.18	0.207	51
STAI Time 1	-0.058	0.66	60
STAI Time 2	0	0.999	60
STAI Time 3	0.031	0.814	60

Discussion

The current study hypothesized that participants in the clary sage group would experience a greater reduction in salivary cortisol presence following the TSST, when compared to the control group which received sweet almond oil. This reduction would exhibit inhalation of the scent of clary sage essential oil having a physiological anxiolytic effect. Preliminary data analysis of saliva samples indicated no significant difference between participants exposed to the scent of clary sage essential oil or sweet almond oil following the TSST and there was no significant difference 40 minutes following the TSST. This finding is consistent with that of

previous literature in which Seol and colleagues (2013) examined the anxiolytic effects of clary sage vapor inhalation 60-minutes during a stressful medical procedure. Clary sage essential oil was not found to significantly reduce salivary cortisol presence (Seol et al., 2013). Based on the current study's results, as well as that of previous literature, clary sage essential has not been found to reduce salivary cortisol in response to acute stress.

The current study also examined blood pressure as another physiological indicator of stress to which potential anxiolytic effects of clary sage essential oil would negatively impact. It was hypothesized that participants in the clary sage group would have a greater reduction in blood pressure following the TSST. Preliminary data analysis revealed no significant differences in systolic blood pressure immediately following the TSST and 20 minutes following the TSST. No significant difference was observed in systolic blood pressure immediately following the TSST and 40 minutes following the TSST as well. Results are similar to that of diastolic blood pressure measures. No significant difference was found in diastolic blood pressure measures immediately following the TSST and 20 minutes following the TSST. Results indicated no significant difference between groups in diastolic blood pressure immediately following the TSST and 40 minutes following the TSST. These results are inconsistent with that of previous literature in which Seol and colleagues (2013) found a significant reduction in participants' diastolic blood pressure when exposed to clary sage vapor. Data collection is currently ongoing in the present study. Once the total estimated sample size has been reached, blood pressure data should be reexamined in order to determine if any significant effects of clary sage essential oil scent on blood pressure are present. At the present point in preliminary data analysis, a definite conclusion cannot be made. Affective indicators of stress will now be discussed.

The current study hypothesized participants exposed to clary sage essential oil would have a reduction in state anxiety levels following scent administration, compared to participants who received sweet almond oil. This was assessed using the STAI-6. Preliminary analysis has suggested no significant difference between the clary sage essential oil and sweet almond oil groups in responses to the STAI-6. Previous literature has not examined the impact of clary sage essential oil scent inhalation on psychological indicators of anxiety. At this time, the hypothesis that clary sage essential oil scent inhalation would reduce state anxiety following an acute stress experience is unsupported.

The current study utilized the DASS21 scale to control for participants' depression scores within the past seven days of study participation. Upon preliminary analysis no significant correlations were determined to exist between the DASS21 depression score, salivary cortisol presence, blood pressure, and responses to the STAI-6. Previous literature has suggested clary sage essential oil injection in rats has an antidepressant like effect and reduces immobility time in the FST task (Seol et al., 2010). Furthermore, Gross and colleagues (2013) examined the chronic administration of clary sage essential oil in mice. Authors found that dominant and submissive mice displayed a reduction in anxiety-like behaviors, dominant mice displayed a decrease in dominant behavior, and submissive mice had reduced corticosterone when given the clary sage essential oil orally. At this time no correlations between participants' depression score and trait anxiety have been found.

The present study examined physiological and psychological indicators of anxiety following the TSST to determine the anxiolytic effects of clary sage essential oil scent inhalation. At this time no significant conclusions can be made. Despite the inconclusive results it is important to note the limitations of the present data.

Data Limitations

Data collection of the total estimated sample size has not yet been reached. Currently data has been collected on 60 participants, while power analysis has suggested a sample size of 102. Given the analysis of the total sample, results may indicate that clary sage essential oil reduces diastolic blood pressure to a significant degree, in comparison to sweet almond oil. The current study is underpowered, further data collection is needed. Several other limitations that must be considered as well, with data collection ongoing.

The current study collected data in one small laboratory room. The scent of clary sage essential oil has remained somewhat constant in the room, despite storing clary sage essential oils in sealed plastic bags, ventilating the room with an oscillating floor fan, and frequently discarding used essential oil cotton pads. This somewhat continuous scent could be acting as a confound. This could be corrected by designating rooms for the testing of clary sage essential oil and sweet almond oil individually.

Data collection has also taken place during winter months. Sinus colds, mucus presence, or other respiratory illnesses more commonly seen in the winter could have impacted observed results. This could be corrected by explicitly screening participants for illness before study participation, and by holding data collection throughout a year.

Future Applications

Given the current results, no anxiolytic effects of clary sage essential oil scent inhalation have been determined. Future testing should include the examination of the form of experimental stressor used to elicit a stress response. Previous research has utilized physical stressors, those observed in medical procedures. The current study utilized the TSST which elicits a stress response based on social evaluative threat (Dickerson & Kemeny, 2004).

Exposure time to the scent of clary sage, as well as the method used to present essential oil scent, may play a role in the extent of anxiolytic effects. The current study exposed participants to the scent of clary sage for five minutes and had participants inhale the scent from one milliliter of essential oil on a cotton pad following the TSST. Previous research by Seol and colleagues (2013) exposed participants to the scent of clary sage essential oil vapor for one hour during a stressor. Exposure time to the scent of clary sage essential oil, as well as method of scent delivery, may elicit different biological processes. The five-minute exposure time to the scent of clary sage essential oil may not have been long enough to elicit potential anxiolytic benefits.

With sufficient power and statistically significant results, this study will give validation to use of clary sage aromatherapy as a secondary treatment for anxiogenic disorders, which has been found to reduce anxiety like behaviors in other mammals (Gross et al., 2013). This study would lay the groundwork for future, more rigorous testing regarding the therapeutic effects of clary sage social stress situations in everyday life, rather than medical settings (Seol et al., 2013).

References

Chen, W., Chen, F., Feng, Y., Chen, A., & Zheng, D. (2017). Quantitative assessment of blood pressure measurement accuracy and variability from visual auscultation method by observers without receiving medical training. *BioMed Research International*, *2017*, Article ID 3537079. https://doi.org/10.1155/2017/3537079

Dickerson, S. S., & Kemeny, M. E. (2004). Acute stressors and cortisol responses: A theoretical integration and synthesis of laboratory research. *Psychology Bulletin*, *130*(3), 355-391. https://doi.org/10.1037/0033-2909.130.3.355

Farrar, A. J., & Farrar, F. C. (2020). Clinical aromatherapy. *Nursing Clinics of North America*, *55*(4), 486-504. https://doi.org/10.1016/j.cnur.2020.06.015.

Faul, F., Erdfelder, E., Lang, A. G., & Buchner, A. (2007). G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods*, *39*(2), 175–191. https://doi.org/10.3758/bf03193146

Fernandes, A., Skinner, M. L., Woelfel, T., Carpenter, T., & Haggerty, K. P. (2013). Implementing self-collection of biological specimens with a diverse sample. *Field Methods*, *25*(1), 58-73. https://doi.org/10.1177/1525822X12453526

Fung, J., & Tsang, H. (2017). Management of behavioral and psychological symptoms of dementia by an aroma-massage with acupressure treatment protocol: A randomized clinical trial. *Journal of Clinical Nursing*, *27*(9-10), 1812-1828. https://doi.org/10.1111/jocn.14101

Ghavami, T., Kazeminia, M., & Rajati, F. (2022). The effect of lavender on stress in individuals: A systematic review and meta-analysis. *Complementary Therapies in Medicine*, *68*, Article ID 102832. https://doi.org/10.1016/j.ctim.2022.102832

Gross, M., Nesher, E., Tikhonov, T., Raz, O., Pinhasov, A. (2013). Chronic food administration of salvia sclarea oil reduces animals' anxious and dominant behavior. *Journal of Medicinal Food*, *16*(3), 216-222. https://doi.org/10.1089/jmf.2012.0137

Hartley, N., & McLachlan, C. S. (2022). Aromas influencing the GABAergic system. *Molecules*, *27*(8), Article ID 2414. https://doi.org/10.3390/molecules27082414

Hellhammer, D. H., Wüst, S., & Kudielka, B. M. (2009). Salivary cortisol as a biomarker in stress research. *Psychoneuroendocrinology*, *34*(2), 163-171. https://doi.org/10.1016/j.psyneuen.2008.10.026

IBM Corp. (2020). IBM SPSS Statistics for Windows (Version 28.0) [Computer software]. IBM Corp

Kim, C., & Song, C. (2022). Physiological and psychological relaxation effects of fir essential oil on university students. *International Journal of Environmental Research and Public Health*, *19*(9), Article ID 5063. https://doi.org/10.3390/ijerph19095063

Kirschbaum, C., Pirke, K. M., & Hellhammer, D. H. (1993). The 'trier social stress test'–a tool for investigating psychobiological stress responses in a laboratory setting. *Neuropsychobiology, 28(1-2), 76-81.* http://doi.org/10.1159/000119004

Kobayashi, H., Song, C., Ikei, H., Park, B.-J., Kagawa, T., & Miyazaki, Y. (2017). Diurnal changes in distribution characteristics of salivary cortisol and immunoglobulin concentrations. *International Journal of Environmental Research and Public Health*, *14*(9), Article ID 987. https://doi.org/10.3390/ijerph14090987

Kudielka, B. M., Hellhammer, D. H., & Kirschbaum, C. (2007). Ten years of research with the trier social stress test--revisited. *Social Neuroscience: Integrating Biological and Psychological Explanations of Social Behavior* (56–83).

Kuo, T. (2017). A study about the inhibition effects of jasmine essential oil on the central nervous system. *Journal of Health Science*, *7*(4), 67-72. http://doi.org/10.5923/j.health.20170704.01

Lopresti, A. L. (2017). Salvia (sage): A review of its potential cognitive-enhancing and protective effects. *Drugs in R&D*, *17*(1), 53-64. https://doi.org/10.1007/s40268-016-0157-5

Lovibond, P. F., & Lovibond, S. H., (1995). The structure of negative emotional states: comparison of the depression anxiety stress scale (DASS) with the beck depression and

anxiety inventories. *Behavior Research and Therapy*, 33(3), 335-343. https://doi.org/10.1016/0005-7967(94)00075-u

Mitic, M., Zrnić, A., Wanner, J., & Stappen, I. (2020). Clary sage essential oil and its effect on human mood and pulse rate: an in vivo pilot study. *Planta medica*, *86*(15), 1125–1132. https://doi.org/10.1055/a-1211-6325

Nuiden, N., Siripornpanich, V., Sayorwan, W., Foto, T. G., Palanuvej, C., & Ruangrungsi, N. (2021). The effects of sweet almond oil inhalation on brainwave activity. *Journal of Public Health and Development*, *19*(1), 65–74. Retrieved from https://he01.tci-thaijo.org/index.php/AIHD-MU/article/view/244269

Salivary cortisol elisa kit. (2024*). Expanded range high sensitivity salivary cortisol enzyme immunoassay kit*. Salimetrics. https://salimetrics.com/wp-content/uploads/2018/03/salivary-cortisol-elisa-kit.pdf

Seol, G., Lee, Y., Kang, P., You, J., Park, M., & Min, S. (2013). Randomized controlled trail for salvia sclarea or lavandula angustifolia: Differential effects on blood pressure in female patients with urinary incontinence undergoing urodynamic examination. *The Journal of Alternative and Complementary Medicine*, *19*(7), 664-670. https://doi.org/10.1089/acm.2012.0148

Seol, G., Shim, H., Kim, P., Moon, H., Lee, K., Shim, I., Suh, S., & Min, S. (2010). Antidepressant-like effects of salvia sclarea is explained by modulation of dopamine activities in rats. *Journal of Ethnopharmacology*, *130*(1), 187-190. https://doi.org/10.1016/j.jep.2010.04.035

Thi Hong Le, M., Duc Tran, T., Holton, S., Thanh Nguyen, H., Wolfe, R., Fisher, J. (2017). Reliability, convergent validity and factor structure of the DASS-21 in a sample of

Vietnamese adolescents. *PLOS ONE*, *12*(7). Article ID e0180557. https://doi.org/10.1371/journal.pone.0180557

Tluczek, A., Henriques, J. B., & Brown, R. L. (2009). Support for the reliability and validity of a six-item state anxiety scale derived from the state-trait anxiety inventory. *Journal of Nursing Measurement*, *17*(1), 19–28. https://doi.org/10.1891/1061-3749.17.1.19

Tsang, H. H., Cheung, W. M., Chan, A. L., Fung, K. T., Leung, A. Y., & Aw, D. H. (2015). A pilot evaluation on a stress management programme using a combined approach of cognitive behavioral therapy (CBT) and complementary and alternative medicine (CAM) for elementary school teachers. *Stress and Health*, *31*(1), 35-43. https://doi.org/10.1002/smi.2522.

Villareal, M. O., Ikeya, A., Sasaki, K., Arfa, A., Neffati, M., & Isoda, H. (2017). Anti-stress and neuronal cell differentiation induction effects of Rosmarinus officinalis L. essential oil. *BMC Complementary and Alternative Medicine*, *17*(1). Article ID 549. https://doi.org/10.1186/s12906-017-2060-1

Appendices

Appendix A: Demographics Survey

Directions: Please answer the following questions to the best of your ability. The purpose of this questionnaire is to help us get general information about you.

1) Which describes you?
   a) Male
   b) Female
   c) Other
2) What is your age in years?
3) What is your ethnicity?
   a) White
   b) Hispanic or Latino

c) Black or African American

d) Native American or American Indian

e) Asian or Pacific Islander

f) More than one ethnicity

g) Other

4) What is your current employment status?

a) Employed full time (40 + hours a week)

b) Employed part time (less than 40 hours a week)

c) Unemployed

d) Student

e) Self-employed

f) Retired

5) What is your overall health?

1 2 3 4 5 6 7 8 9 10

Poor Excellent

6) What is your overall health today?

1 2 3 4 5 6 7 8 9 10

Poor Excellent

Salivary Cortisol Collection

7) Have you consumed alcohol in the past 12 hours?

a) Yes

b) No

8) Have you eaten or drank anything in the past hour? This includes caffeine products.

a) Yes

b) No

9) Have you had a recent infection?

a) Yes

b) No

10) Have you had any social problems (problems related to other people) in the past 3 hours?

a) Yes

b) No

Female Participants

11) When was your last menstrual period? _____

12) Are you currently pregnant or think you may be pregnant?

a) Yes

b) No

13) Are you currently taking hormonal birth control?

a) Yes

b) No

Appendix B: Institutional Review Board Approval Letter

To:
CaSandra Stanbrough
Psychology

RE: Notice of IRB Approval
Submission Type: Initial
Study #: IRB-FY2024-91
Study Title: Aromatherapies and Stress: An Examination of Salvia Sclarea in Acute Stress Conditions
Decision: Approved

Approval Date: October 2, 2023

This submission has been approved by the Missouri State University Institutional Review Board (IRB). You are required to obtain IRB approval for any changes to any aspect of this study before they can be implemented. Should any adverse event or unanticipated problem involving risks to subjects or others occur it must be reported immediately to the IRB.

___
This study was reviewed in accordance with federal regulations governing human subjects research, including those found at 45 CFR 46 (Common Rule), 45 CFR 164 (HIPAA), 21 CFR

50 & 56 (FDA), and 40 CFR 26 (EPA), where applicable.

Researchers Associated with this Project:
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