



Original Article

Effects of nighttime lavender aromatherapy on mood and physiological indices of stress in healthy young females

KANAKO SAZAWA, BS¹⁾, SHIGEO KAWADA, PhD¹⁾, YOSHIKO OGAWA, MD, PhD^{1)*}

¹⁾ Department of Sport and Medical Science, Faculty of Medical Technology, Teikyo University: 359 Otsuka, Hachioji-shi, Tokyo 192-0395, Japan

Abstract. [Purpose] Inhalational aromatherapy using lavender essential oil or essence is known to alleviate pain and anxiety during rehabilitation. However, the effects remain unclear in individuals who are unaware of their pain and anxiety. In this study, we investigated the effects of lavender aromatherapy during sleep in females who did not experience pain or anxiety. [Participants and Methods] The study included 24 healthy females who were randomly allocated to control and aromatherapy groups. The control group used skin patches without aroma, and the aromatherapy group used lavender aroma-infused skin patches for seven consecutive nights. Psychological and physiological indices were measured before, during, and after the intervention. [Results] The lavender aroma-infused skin patches ameliorated a negative mood associated with fatigue and anxiety. However, neither group showed a change in pulse rate and salivary cortisol concentration upon waking. Furthermore, no significant intergroup difference was observed in sleep quality. [Conclusion] Lavender aromatherapy during sleep improved a negative mood associated with fatigue and anxiety in females who did not experience pain and anxiety; however, physiological indices remained unaffected.

Key words: Profile of mood states, Sleep quality, Cortisol

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INTRODUCTION

Aromatherapy through inhalation of aromas, which uses essential oils from plants, is widely used to alleviate pain, anxiety, and sleep disorders in a variety of diseases and injuries^{1–4)}. Targets of aromatherapy also vary considerably, from elderly people with cardiovascular diseases to athletes with orthopedic injuries^{3, 5)}. The effects of aromatherapy have been theorized to result from the binding of chemical components of essential oils to receptors in the olfactory bulb, affecting the limbic system, which is the emotional center in the brain⁶⁾. As basic data and case reports on aromatherapy have increased, it is now being used in the medical field. In fact, lavender, one of the plants widely used in aromatherapy, is reported to be helpful in alleviating depression, pain, and anxiety in patients undergoing abdominal surgery⁷⁾ and in females undergoing cesarean section⁸⁾.

However, in intervention studies of aromatherapy, the participants are mostly those with pain, anxiety, or sleep disorders. The effects of aromatherapy in individuals who are unaware of such symptoms are not fully understood. For aromatherapy to be used safely in a wide range of medical fields, it is necessary to consider the effects of inhalation of aroma in individuals who do not experience pain or other subjective symptoms. Thus, the present study aimed to determine whether nighttime aromatherapy affects mood and physiological indices of stress in healthy young females.

*Corresponding author. Yoshiko Ogawa (E-mail: yogawa@main.teikyo-u.ac.jp)

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PARTICIPANTS AND METHODS

All experimental procedures were performed in accordance with the guidelines of the Declaration of Helsinki and were approved by the Ethics Committee of the Teikyo University (approval no. 18-050). The participants were informed regarding the experimental procedures, risks, and purpose of the study and signed an informed consent before undergoing the experimental procedures. Twenty-four healthy young females participated in this study. It has been reported that 50% or more of Japanese young people suffer from excessive daytime sleepiness⁹). The rate of difficulty falling asleep and midway awakening in young females is reportedly approximately twice as high as in males¹⁰). Since sleep disturbances also lead to fatigue and anxiety, young females were recruited as participants for this study. The inclusion criterion for this study was healthy females aged over 18 years. The exclusion criteria were as follows: pregnant females, lactating mothers, smokers, females who worked on night-shift duty, females who used hypnotics, females who were allergic to lavender, females with a history of adverse events associated with aromatherapy, and females who have experienced pain or anxiety. This study was a randomized, controlled, single-blind trial. The participants were randomly allocated to either an aromatherapy (AROM) group (n=12; age, 20.1 ± 1.3 years; height, 160.8 ± 6.6 cm; body weight, 55.2 ± 7.0 kg) or a control (CONT) group (n=12; age, 20.4 ± 1.5 years; height, 160.4 ± 6.9 cm; body weight, 53.3 ± 7.0 kg). The participants were instructed to place a skin patch under their clothing just before going to bed at night and to sleep as usual at home for seven consecutive nights. The AROM group used commercial lavender aroma-infused skin patches (Aroma Patch[®]; Japan Detox, Toyama, Japan), whereas the CONT group used skin patches without aroma (EM One-Touch Pad[®]; New Tac Kasei Co., Ltd., Kagawa, Japan). These patches were removed and discarded once the participant was awake. In this study, we used lavender, which is one of the aromas reported to relieve pain and anxiety and is relatively easy to obtain. Mood assessment and saliva collection were conducted between 8:30 am and 10:00 am before the intervention (day 0), in the morning after the first intervention night (day 1), and after the intervention (day 7). Pulse rate was measured by the participants themselves upon waking up before, during, and after the intervention; in particular, they were instructed to measure the pulse rate of the radial artery for 60 seconds immediately after waking up. The participants were asked to refrain from other aromatherapies during their participation in the study.

Mood was assessed using the Profile of Mood States 2nd Edition (POMS 2). The POMS 2, which is a modified version of the POMS, simultaneously evaluates seven moods: anger-hostility (AH), confusion-bewilderment (CB), depression-dejection (DD), fatigue-inertia (FI), tension-anxiety (TA), vigor-activity (VA), and friendliness (F). The total mood disturbance (TMD) score was calculated using the following formula:

$$\text{TMD score} = (\text{AH} + \text{CB} + \text{DD} + \text{FI} + \text{TA}) - \text{VA}$$

We used the Japanese version of short-form POMS 2, which includes 35 questions, to reduce the burden of assessment on the participants¹¹).

Saliva was collected using a cotton swab (Salivette[®]; SARSTEDT AG & CO., Numbrecht, Germany). The participants were not allowed to drink alcoholic beverages on the night before saliva collection. Additionally, eating, drinking (except for water), exercising, and bathing in the morning of saliva collection were prohibited. After rinsing the mouth thoroughly with water, the participants were instructed to gently chew the swab for 1 min to allow the swab to absorb the saliva. The saliva-saturated swab was placed in a tube and centrifuged at 3,000 rpm for 5 min. Obtained saliva samples were frozen at -80°C until the time of analysis. Salivary cortisol concentration was measured using an enzyme-linked immunosorbent assay kit (Salimetrics, LLC, Carlsbad, CA, USA) according to the manufacturer's protocol.

Pre-intervention sleep quality was assessed using the Pittsburgh Sleep Quality Index¹²) for Japanese (PSQI-J)^{13, 14}). Sleep disorder was considered to be present when an individual's score was ≥ 6 ^{12, 14}). Because the PSQI-J is designed to investigate sleep quality over the past month, it is not suitable for sleep quality evaluation before and after a short-term intervention experiment. Therefore, the Ogrī-Shirakawa-Azumi sleep inventory MA version (OSA-MA)¹⁵), which is popularly used for sleep quality evaluation in Japan, was adopted to investigate changes in sleep quality before and after the intervention. The OSA-MA evaluates five factors of sleep quality—namely, sleepiness on rising, initiation and maintenance of sleep, frequent dreaming, refreshing, and sleep length. Scores were calculated using an MS Excel spreadsheet¹⁵), with higher scores being indicative of good sleep quality. The average amount of sleep (hours) during the intervention (day 0 to day 7) was assessed using the participants' self-reported data.

We asked the participants whether they perceived any aroma on the patches. If they answered, "Yes", we determined their preference for the aroma on a scale of 1 to 5, with 1="I like the aroma very much", 2="I like the aroma a little", 3="I do not like or dislike the aroma", 4="I hardly like the aroma", and 5="I do not like the aroma".

Data are presented as mean ± standard deviation (SD). Data on the participants' characteristics were analyzed using the unpaired t-test or χ^2 test. The POMS 2 scores, salivary cortisol concentration, pulse rate, and OSA-MA scores were analyzed using a two (condition [with/without aromatherapy]) × three (time of measurement [before the intervention, in the morning after the first intervention night, and after the intervention]) analysis of variance (ANOVA) to determine the interaction and main effects. When statistical significance was determined by ANOVA, Bonferroni's post-hoc test was used to compare the

data. Statistical analyses were performed using SPSS Statistics version 25.0 (IBM, Tokyo, Japan). Two-tailed-p-values of <0.05 were considered indicative of statistical significance.

RESULTS

Fifty percentage or more participants in the CONT and the AROM groups were diagnosed with a sleep disorder based on the PSQI-J scores (Table 1). The incidence of sleep disorder in the present study is similar to that in a previous study that demonstrated sleep problems in Japanese young males and females⁹). The pre-intervention sleeping hour was similar between the groups (Table 1).

The baseline POMS 2 scores are shown in Table 2. Pre-intervention (day 0) TMD and POMS 2 subscale scores were not significantly different between the groups. Regarding the chronological TMD scores, two-way ANOVA showed a statistical difference in time ($p<0.01$); however, no interaction between time and condition was detected. After Bonferroni's test, the TMD scores on days 1 ($p<0.01$) and 7 ($p<0.05$) were significantly lower than those on day 0 in the AROM group; on the other hand, no significant difference in TMD scores was observed over time in the CONT group. Concerning the FI score as an index of fatigue-inertia, two-way ANOVA showed an interaction between time and condition ($p<0.05$) and a statistical difference in time ($p<0.01$). After Bonferroni's test, the FI score on day 7 was significantly lower ($p<0.01$) than that on day 0 in the AROM group, but there was no significant difference in FI scores over time in the CONT group. Regarding the AH score as an index of anger-hostility, two-way ANOVA showed a statistical difference in time ($p<0.05$), but no interaction between time and condition was detected. After Bonferroni's test, the AH score on day 7 was significantly lower ($p<0.05$) than that on day 0 in the AROM group; in contrast, there was no significant difference in AH scores over time in the CONT group. For the CB, DD, and TA scores as indices of confusion-bewilderment, depression-dejection, and tension-anxiety, two-way ANOVA showed no significant difference in time, and no interaction between time and condition was noted. For the VA and F scores as indices of vigor-activity and friendliness, two-way ANOVA showed no significant difference in time, and no interaction between time and condition was observed.

Chronological changes in cortisol concentration are shown in Table 3. For salivary cortisol concentration, two-way ANOVA showed no significant difference in time, and no interaction between time and condition was found.

Table 1. Sleep assessment prior to the intervention

	CONT (n=12)	AROM (n=12)
PSQI-J (scores)	5.7 ± 3.0	5.8 ± 3.2
Sleep disorder (n)	6 (50.0%)	7 (58.3%)
Sleeping hour (hours)	6.5 ± 0.7	6.7 ± 0.8

CONT: control group; AROM: aromatherapy group; PSQI-J: the Pittsburgh Sleep Quality Index for Japanese.

Values, except sleep disorder, are expressed as mean and SD. Value of sleep disorder is expressed as number of participants (rate).

Sleep disorder is defined as PSQI-J score ≥6.

The average of sleep hours was calculated by self-reported data during intervention period (from day 0 to day 7).

Table 2. Chronological changes in the POMS 2 score

	CONT (n=12)			AROM (n=12)		
	Day 0 (pre)	Day 1	Day 7 (post)	Day 0 (pre)	Day 1	Day 7 (post)
TMD	45.6 ± 8.0	43.3 ± 6.1	42.8 ± 4.6	45.8 ± 5.7	42.8 ± 5.6**	41.4 ± 7.3*
AH	43.8 ± 7.8	39.3 ± 3.1	40.4 ± 3.5	43.1 ± 6.9	41.2 ± 5.5	40.1 ± 4.5*
CB	48.5 ± 10.5	44.7 ± 6.8	42.1 ± 4.6	46.3 ± 6.6	44.3 ± 8.3	43.6 ± 8.7
DD	46.6 ± 5.7	45.1 ± 6.2	43.9 ± 4.0	46.6 ± 7.7	45.2 ± 6.0	45.3 ± 7.9
FI	45.3 ± 7.6	43.0 ± 6.9	43.3 ± 6.3	49.2 ± 5.7	44.5 ± 5.9	40.4 ± 4.8**
TA	45.3 ± 10.9	42.4 ± 7.6	40.4 ± 4.6	44.7 ± 7.8	41.7 ± 7.8	42.6 ± 9.5
VA	47.0 ± 7.4	44.9 ± 7.9	47.1 ± 11.4	50.0 ± 9.4	50.4 ± 9.0	52.8 ± 9.9
F	49.8 ± 9.2	47.6 ± 9.6	46.3 ± 12.3	53.0 ± 7.5	53.3 ± 8.8	52.2 ± 10.6

AH: anger-hostility; AROM: aromatherapy group; CB: confusion-bewilderment; CONT: control group; DD: depression-dejection; F: friendliness; FI: fatigue-inertia; POMS 2: the Profile of Mood States 2nd Edition; TA: tension-anxiety; TMD: total mood disturbance; VA: vigor-activity.

Values are expressed as mean and SD.

* $p<0.05$, ** $p<0.01$ compared with the score on day 0 in the AROM group.

Chronological changes in the pulse rate from day 0 to day 7 are presented in Table 4. Pre-intervention pulse rate was not significantly different between the groups. Further, two-way ANOVA showed no significant difference in pulse rate in time and in condition.

Chronological changes in the OSA-MA scores are shown in Table 5. With respect to the scores for all five factors, two-way ANOVA showed no significant difference in time. No interaction between time and condition was noted.

All participants in the AROM group responded that they perceived the lavender aroma. Contrarily, all participants in the CONT group responded that they did not perceive the lavender aroma. Regarding the preference for the aroma, 1, 2, 3, 4, and 5 participants answered with 1, 3, 4, 3 and 1, respectively, on a scale of 1 to 5. No adverse effects were observed during the study period.

DISCUSSION

The use of lavender aroma-infused skin patches during sleep improved TMD scores and the scores of two negative subscales of POMS 2 (AH and FI) over time in healthy young females. Since TMD, an evaluation of overall negative mood, was improved, the aroma patch in this study may have improved the mood of participants who did not experience pain and anxiety. In particular, for FI as an index of fatigue-inertia, an interaction was detected between time and condition, and FI was significantly reduced compared to baseline, after 7 days of nighttime aromatherapy. This result suggests that using lavender aroma-infused skin patches during sleep significantly improved fatigue and inertia in healthy young females. To our knowledge, this is the first report about the effects of nighttime lavender aromatherapy on mood in healthy young females who did not experience pain and anxiety. On the other hand, no interactions between time and condition were detected for TMD and AH (anger-hostility) because there were slight placebo effects in the CONT group, although these scores were significantly reduced in the AROM group, compared to baseline, after 7 days of nighttime aromatherapy. A similar placebo effect on acute stress was shown in healthy young adults receiving aromatherapy¹⁶). In addition, the effect of lavender aromatherapy on psychological indices of relaxation were considered to have been related to the participants' expectations, not to the aroma, in healthy young females¹⁷). The results of present study, which was single-blind, may have been associated with expectancy biases, although all participants in the CONT group mentioned that they did not perceive the lavender aroma. The implementation of overnight aromatherapy using lavender oil patches, similar to the present study, improved sleep quality and duration and also reduced anxiety in inpatients in a hematology-oncology unit¹⁸). Another study reported that aromatherapy improved

Table 3. Chronological changes in salivary cortisol concentration

(µg/dL)	Day 0 (pre)	Day 1	Day 7 (post)
CONT (n=12)	1.23 ± 0.47	1.34 ± 0.43	1.35 ± 0.36
AROM (n=12)	0.92 ± 0.50	1.03 ± 0.35	1.18 ± 0.36

CONT: control group; AROM: aromatherapy group.
Values are expressed as mean and SD.

Table 4. Chronological changes in pulse rate

(bpm)	Day 0 (pre)	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7 (post)
CONT (n=12)	62.3 ± 10.9	56.9 ± 5.5	56.7 ± 6.5	58.8 ± 7.7	56.8 ± 5.7	56.7 ± 7.5	59.3 ± 7.5	57.0 ± 4.6
AROM (n=12)	63.5 ± 7.0	61.3 ± 7.9	63.2 ± 6.9	64.8 ± 7.2	61.2 ± 7.2	61.5 ± 7.3	61.0 ± 7.4	63.5 ± 9.4

CONT: control group; AROM: aromatherapy group.
Values are expressed as mean and SD.

Table 5. Chronological change the OSA-MA score

	CONT (n=12)			AROM (n=12)		
	Day 0 (pre)	Day 1	Day 7 (post)	Day 0 (pre)	Day 1	Day 7 (post)
Sleepiness on rising score	14.4 ± 4.8	15.1 ± 6.7	19.2 ± 6.9	19.0 ± 6.4	19.5 ± 4.5	19.1 ± 7.0
Initiation and maintenance of sleep score	18.6 ± 6.1	17.3 ± 7.7	21.1 ± 3.7	20.9 ± 4.2	22.2 ± 3.6	20.9 ± 5.1
Frequent dreaming score	22.5 ± 9.0	23.9 ± 8.3	25.2 ± 7.3	24.8 ± 5.9	23.0 ± 8.4	22.1 ± 10.0
Refreshing score	12.2 ± 4.9	14.2 ± 4.7	16.1 ± 7.4	17.6 ± 6.0	17.0 ± 3.4	15.1 ± 4.4
Sleep length score	17.2 ± 4.8	18.3 ± 6.7	19.5 ± 7.1	20.7 ± 6.6	19.9 ± 9.0	20.2 ± 8.9

CONT: control group; AROM: aromatherapy group; OSA-MA: the Ogri-Shirakawa-Azumi sleep inventory MA version.
Values are expressed as mean and SD.

sleep quality in healthy males and females; however, the participants were informed in advance that the aroma oil used in the study had a calming effect and improved sleep quality¹⁹). As observed in the CONT group of our study, aromatherapy has a slight placebo effect, so the discrepancy between our results and theirs may have been due to differences in experimental procedures. Furthermore, although 50% or more of the participants in the present study had sleep disorder, the POMS 2 scores of the majority of the participants were within the normal range. It implies that the degree of sleep disorder observed in the present study was not as severe as those observed in patients with anxiety. In addition, no significant intergroup difference was observed in sleep quality assessed by the OSA-MA. Thus, since many factors may influence the effects of the lavender aroma on POMS 2 and sleep quality, aromatherapy may have not been appreciable to fully outperform the CONT group in the present healthy young females. Furthermore, nighttime aromatherapy using lavender aroma-infused skin patches, at least, did not aggravate the mood in healthy young females, regardless of whether they liked lavender aroma or not.

In the present study, salivary cortisol concentration was measured as an index of stress response via the hypothalamic-pituitary-adrenal (HPA) axes. No significant intergroup difference was observed in salivary cortisol concentration in the present study. A few studies which examined the acute effect of lavender aroma inhalation reported that salivary cortisol level did not change in the healthy young adults^{20, 21}); however, no study has reported the effect of repeated lavender aroma inhalation on salivary cortisol level. The results of the present study suggest that lavender aroma may not affect stress response via the HPA axis in healthy young females. In addition, the baseline states of the participants may have affected the physiological indices. Further studies are needed to demonstrate the beneficial effects of nighttime aromatherapy on physiological stress responses in participants with high stress.

There were also no significant differences in pulse rate in the AROM and CONT groups. Heart rate is modulated by cardiac autonomic nervous activity, and reduction in heart rate reflects a reduction of sympathetic nervous activity and/or the enhancement of parasympathetic nervous activity. Lavender, especially its linalool component, affects the autonomic nervous system, inhibits sympathetic nervous activity, and excites parasympathetic nervous activity²²). Lavender aromatherapy was shown to decrease heart rate in patients with cardiovascular disease^{2, 23}). However, in healthy individuals, changes in heart rate caused by the inhalation of the lavender aroma were inconsistent^{24, 25}). Thus, the effect of the lavender aroma on heart rate may be more profound in patients with disorders of the autonomic nervous system than in healthy adults.

The present study had a few limitations. First, the sample size was small. Second, the participants were young females. These factors may limit the generalizability of the findings of the present study. Despite these limitations, the results of the present study suggest that aromatherapy during sleep may be useful for stabilizing mood in young females who do not experience pain or anxiety. Further studies are needed to demonstrate the beneficial effects of nighttime aromatherapy on mood in various populations.

In conclusion, the use of lavender aroma-infused skin patches during sleep partly improved mood in young females who did not experience pain or anxiety. The result of the present study indicates that nighttime aromatherapy using lavender aroma-infused skin patches is an easy, inexpensive, and safe approach that can stabilize the mood of an individual.

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Conflict of interest

The authors report no conflicts of interest.

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