











## ORIGINAL ARTICLE

# Fragrance preference of essential oil blends for reducing stress and sleep problems in adult women

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## ABSTRACT

### BACKGROUND

Aromatherapy with a blend of essential oils is an alternative treatment for stress and sleep disturbances, providing stress relief and sleep enhancement, while increasing the efficacy and scent appeal. This study aimed to formulate an essential oil blend with the potential to reduce stress and enhance sleep quality, and to identify the most preferred blend based on participant satisfaction.

### METHODS

A cross-sectional study was conducted involving 63 female volunteers aged 25 - 50 years. Three formulations (A, B, and C) of essential oil blends were developed, each with a distinct scent profile—floral, unisex, and aromatic. A preference test was conducted using a sensory evaluation method, assessing participants' satisfaction of the blends through inhalation. Preferences were quantified using a structured scoring system. Formula B was the most popular and was further refined into two recipes, B1 and B2. B2 emerged as the preferred option, and its primary component was identified using gas chromatography-mass spectrometry (GC-MS).

### RESULTS

Formulation B, rich in bergamot oil, received significantly higher hedonic ratings than Formulation A ( $p < 0.05$ ) and was the most preferred in interviews. Participants recommended a milder, more balanced scent for relaxation and sleep. While B1 and B2 showed no significant differences, B2 was chosen for its higher bergamot content, fresher unisex profile, and reduced floral notes. GC-MS analysis identified limonene, menthol, and linalool as its main components.

### CONCLUSION

This study demonstrated that an essential oil blend predominantly composed of bergamot oil was most favored by the volunteers, indicating its potential benefits for promoting relaxation.

**Keywords:** Essential oil blend; sleep; sleep problem; stress; preference, women

## INTRODUCTION

Stress is an adaptive mechanism that helps restore body balance, with the hypothalamic-pituitary-adrenal (HPA) axis playing a key role in regulating physiological processes such as the sleep-wake cycle.<sup>(1)</sup> Stress can negatively impact physical and mental health. This multifaceted phenomenon affects numerous systems, including the immune and reproductive systems, resulting in various physical and psychological symptoms that diminish the quality of life. Stress can be categorized into two types: acute stress, which is triggered by specific events, and chronic stress, which arises from ongoing situations and is associated with various medical conditions.<sup>(2)</sup>

Sleep is essential for maintaining physical health, mental well-being, memory, and learning, and plays a vital role in overall functioning and quality of life. Sleep deprivation has a negative impact on the body, leading to fatigue, daytime drowsiness, and impaired cognitive function.<sup>(3)</sup> Sleep quality pertains to the depth and restorative aspects of sleep, encompassing factors such as ease of falling asleep, total sleep duration, frequency of nighttime awakenings, and overall feeling upon waking in the morning.<sup>(4)</sup> The relationship between stress and sleep is complex but negative, with stress from major life events and daily hassles disrupting normal sleep function. Sleep reactivity refers to the extent to which a person's sleep is disrupted by stress, manifesting as difficulty falling or staying asleep. Research studies have shown that genetics, family history of insomnia, female sex, and environmental stress determines how the sleep system responds to stress.<sup>(5)</sup> Sleep disorders can lead to increased appetite, higher caloric intake, and elevated pro-inflammatory cytokines, blood pressure, insulin, and blood glucose levels. Additionally, sleep deprivation disrupts the neuroendocrine stress response by increasing sympathetic tone and cortisol levels.<sup>(6)</sup> Several studies in both animals and humans have shown that stress can significantly affect the sleep-wake cycle, with varying impact based on the type of stress and whether exposure is acute or chronic.<sup>(7)</sup> Stress can lead to poor sleep and weakened immune function, and sleep problems are closely linked to stress and often associated with depression. Stress-related sleep disturbances can reduce immune function.<sup>(8)</sup>

Cortisol, catecholamines, amylase, and secretory immunoglobulin A (s-IgA) are commonly recognized as stress markers, reflecting mental stress. These markers are closely linked to the autonomic nervous and immune systems. s-IgA continuously decreases in the presence of long-term stress loading.<sup>(9)</sup>

Essential oils act as bioactive agents that influence the nervous system via the olfactory and cutaneous pathways, affecting emotions, memories, mood, and behavior, and interact with the nervous system through two primary pathways: olfactory and cutaneous.<sup>(10)</sup> Inhalation of essential oils activates olfactory receptors in the nasal mucosa, sending electrical signals to the olfactory bulb, which in turn connects to the limbic system, the brain region associated with emotions, memory, mood, learning, and behavior. This interaction allows essential oils to influence emotional states, stress levels, anxiety, depression, motivation, attention, concentration, creativity, and other psychological factors. The cutaneous pathway involves applying essential oils to the skin, typically diluted in a carrier oil, which enables them to penetrate tissues and enter the bloodstream. This results in physiological effects, such as relaxation or stimulation of muscles, nerves, blood vessels, glands, and various organ systems, including the immune, endocrine, digestive, respiratory, and reproductive systems.<sup>(2,3)</sup> Aromatherapy is recognized by the World Health Organization (WHO) as an effective treatment for relieving pain, improving mood, and promoting relaxation, particularly for addressing anxiety and daily stress issues.<sup>(11)</sup> Aromatherapy involves the use of essential oils for treatment, typically through inhalation or topical application.<sup>(12)</sup> Sleep disorders are becoming increasingly common and have a significant impact on health and well-being. Among the various therapeutic options available, essential oils derived from plants are emerging as a promising alternative for improving sleep quality.<sup>(4)</sup>

Various aromatic plants, producing essential oils, are tropical trees native to Southeast Asia, including Thailand, where especially plants that produce bergamot, vetiver, and ylang ylang oils are widely cultivated and used in aromatherapy.<sup>(13,14)</sup> Several essential oils exhibit neuroactive properties that support stress

reduction, cognitive enhancement, mood regulation, and improved sleep. Peppermint oil stimulates the CNS and aids memory and alertness, while ylang ylang oil promotes relaxation through serotonergic and dopaminergic pathways.<sup>(15)</sup> Eucalyptus oil (from *Eucalyptus globulus*) enhances cognition via acetylcholinesterase inhibition. Bergamot oil improves sleep quality and alertness upon waking.<sup>(16)</sup> Chamomile essential oil (from *Matricaria chamomilla* L.) shows potential against stress and anxiety, improves sleep quality, with no adverse events being reported in any of the studies.<sup>(17)</sup> Blending these essential oils helps maximize benefits and obtain satisfactory scents. When blending essential oils, it is important to consider factors such as synergy, affinity, complementarity, harmony, and the unique identity of each oil. A practical approach is to follow the rule of aromatic notes, which suggests mixing oils of different notes for a balanced and lasting aroma, typically using a ratio of approximately 30% high-note oils, 50% middle-note oils, and 20% low-note oils.<sup>(5)</sup> Their application, whether through inhalation or topical use, provides beneficial physiological and psychological outcomes.

With the current emphasis on individualized treatment, the variety of available products, including diffusers and creams, allows for flexible and personalized use. In the clinical settings, incorporation of essential oils into stress management strategies is supported by their ability to regulate emotional responses and induce relaxation. Essential oils provide an effective and customizable approach to stress management, promoting physical and emotional balance.<sup>(6)</sup> A study conducted in Korea revealed that aroma preferences varied significantly depending on the participants' level of education and sleep habits. In particular, high school graduates tended to prefer the scent of rosemary, the sleep-type group favored lemon and lavender scents, but interestingly the insomnia group also preferred the scent of rosemary.<sup>(18)</sup> The development of essential oil blends allows for the inherent benefits of individual oils to be coupled together, potentially resulting in a synergistic effect that exceeds the efficacy of each oil used alone. In practice, aromatherapists seldom rely on single essential oils, instead favoring blends to effectively integrate and enhance their therapeutic properties.<sup>(19)</sup>

Previous research has predominantly focused on single molecules of essential oil components rather than blended formulations,<sup>(20)</sup> thereby overlooking the potential synergistic effects that may enhance therapeutic outcomes when multiple oils are combined. Furthermore, these studies typically did not incorporate a preference test prior to formulation to assess participants' olfactory satisfaction. In contrast, the present study conducted a preference test as an initial step, allowing for the selection and adjustment of the essential oil blend based on volunteer preferences.<sup>(15)</sup> The findings of a clinical study investigating the effects of essential oils and their differential efficacy have not been clearly established. The study utilized a single essential oil without conducting prior preference testing among participants, potentially influencing the therapeutic outcomes due to individual differences in olfactory perception and response.<sup>(21)</sup> Additionally, the key active aromatic constituents of the selected blend were identified through gas chromatography-mass spectrometry (GC-MS) analysis. The objectives of the present study were to formulate an essential oil blend with the potential to reduce stress and enhance sleep quality, and to identify the most preferred blend based on participant satisfaction.

## METHODS

### Research design

A cross-sectional study was conducted at Srinakharinwirot University, Bangkok, Thailand, over a six-month period from January to June 2022.

### Research subjects

63 female volunteers aged 25-50 years, 52 were selected to participate in this study, based on the sample size calculation according to Yamane:<sup>(22)</sup>

$$n = N/(1+Ne^2)$$

where n = sample; N = population; e = error (0.05), resulting in  $n = 60/(1+60(0.05)^2)$  or n=52 persons.

Based on the sample size calculation, a minimum of 52 participants was required for the preference test. Therefore, in the subsequent phase of the study, a total of 63 participants were included to enhance the reliability and confidence in the preference assessment results. Our research study on scent satisfaction for essential oil blends tested the participants' preferences by dropping the essential oil sniff formulation directly on a

scent paper strip. The total number of participants comprised 63 students from Srinakharinwirot University (SWU).<sup>(22)</sup> The volunteers received scent paper strips in 3 formulation and tested these by smell. After that, a scent satisfaction questionnaire classified by hedonic scaling test and strength test was used to evaluate the scent satisfaction of each volunteer. Inclusion criteria: females, having no olfactory disorders, nonsmokers, having no drug abuse. The exclusion criteria included olfactory diseases, a history of cardiovascular or respiratory diseases, pregnancy, lactation, or menstruation.

### Chemicals and materials

Vetiver oil (from *Chrysopogon zizanioides*) was purchased from Royal project, Chiang Mai, Thailand. Bergamot oil (*Citrus bergamia*) and peppermint oil (*Mentha piperita*) were purchased from Botanicessense Essential Oil, Bangkok, Thailand. Eucalyptus oil (*Eucalyptus globulus*), chamomile oil (*Chamomilla recutita*), ylang ylang oil (*Cananga odorata*), lavender oil (*Lavandula angustifolia*), marjoram oil (*Origanum majorana*), and cedarwood oil (*Cedrus atlantica*) were purchased from Devika Enterprise Company Limited, Bangkok, Thailand. The sleep balm formulation and samples were supplied by Cosmed and Anti-Aging Company Limited, Bangkok, Thailand.

### Essential oil blend formulation

Essential oils known for their relaxation and sleep-enhancing properties, as reported in previous studies, were chosen for use in this research. According to a study by Qneibi et al.<sup>(23)</sup> on the pharmacological activities of essential oils

in neurological disorders, and a bibliometric study and visualization analysis by Cao et al.,<sup>(24)</sup> combining multiple essential oils with similar benefits is more effective for promoting relaxation and reducing stress than using a single type. Bergamot oil, peppermint oil, eucalyptus oil, chamomile oil, ylang ylang oil, lavender oil, marjoram oil, vetiver oil, and cedarwood oil were selected to formulate an essential oil blend to reduce stress and sleep problems in our volunteers (Table 1). Three essential blend formulations (A, B, and C) were created with different scent characteristics: Formula A was designed for refreshment and energization, Formula B for floral notes and Formula C for warmth and freshness. All formulations included bergamot oil, ylang ylang oil, and chamomile oil, but there were differences in the proportions of peppermint, eucalyptus, lavender, marjoram, cedarwood, and vetiver oils. Table 1 presents a list of essential oils beneficial for stress and sleep issues, along with notes on each oil based on their evaporation rates.

Among the three essential oil blends, Formula B was the most preferred by the volunteers participating in this study and was further developed into two variations, B1 and B2, each displaying the specific ratio of essential oils listed in Table 2. Another preference test, performed again with the same volunteers, was conducted to compare formulations B1 and B2, in which the concentration of the essential oils was adjusted based on volunteer feedback to enhance the overall satisfaction with formulation B. Ultimately, B2 was selected as the preferred blend, and its main component was identified through gas chromatography-mass spectrometry (GC-MS).

Table 1. Formulation of essential oil blends

List of essential oils for stress with sleep problems	Notes of essential oils based on rate of evaporation	%		
		A	B	C
Bergamot oil		40	41.38	49.02
Peppermint oil	Top note	0	10.34	0.00
Eucalyptus oil		0	0.00	9.80
Chamomile oil		8	2.76	1.96
Ylang ylang oil	Middle note	8	2.76	9.80
Lavender oil		0	0.00	19.61
Marjoram oil		40	41.38	0.00
Vetiver oil	Base note	0	1.38	9.80
Cedarwood oil		4	0.00	0.00
Total		100	100.00	100.00

Table 2. The development of essential oil blend formulation from the preference test

Essential oil blend formulation for stress with sleep problems	% B1	% B2
Bergamot oil	49.02	58.14
Peppermint oil	24.51	17.44
Marjoram oil	9.80	11.63
Ylang ylang oil	9.80	5.81
Chamomile oil	4.90	5.81
Vetiver oil	1.96	1.16
Total	100.00	100.00

The mean percentage of each essential oil included in all formulations was analyzed to identify the predominant aromatic compounds. The results highlighted the key differences in the scent profiles of the two formulations, with the major aroma constituents serving as distinguishing factors that contribute to the unique olfactory characteristics of each blend.

### Preference test

After inhaling a sample of the essential oil blend on the scent paper strips, the volunteers were asked to complete a preference test questionnaire. All parameters of hedonic scale, strength, and satisfaction were rated on a 5-point Likert scale. A hedonic scale can be used to determine the degree of acceptance of one or more products. This scale is a category-type scale with a number of categories ranging from “dislike extremely” to “like extremely”, and includes a neutral midpoint. The participants rated the products on a scale based on their responses. Sensory preference tests are commonly structured to include two key components: hedonic evaluation and intensity assessment. Odor hedonic perception—referring to the degree of pleasantness or unpleasantness associated with a scent—is considered one of the primary and most salient dimensions in olfactory processing. In parallel, the intensity test evaluates the perceived strength of the odor, providing insight into how strongly or weakly the scent is detected by participants. Together, these dimensions offer a comprehensive understanding of olfactory preferences and perception.<sup>(25,26)</sup>

The strength test can be evaluated subjectively in terms of intensity (strength), which is determined by describing the odor or comparing the sample odor to familiar odors.<sup>(27)</sup> Moreover, a preference scale was used to measure satisfaction. The statement may be positive or negative. Usually, a 5-point scale of satisfaction such as the following is used, as shown in Table 3.

Table 3. Blend satisfaction scores

Level	Score	Meaning
5	4.21-5.00	Excellent
4	3.41-4.20	Good
3	2.61-3.40	Moderate
2	1.81-2.60	Low
1	1.00-1.80	Very low

### Gas chromatography mass spectrometry (GC-MS)

The components of the essential oil blend were identified using gas chromatography-mass spectrometry (GC-MS). The results were analyzed based on a test report of the essential oil blends from the Expert Center of Innovative Herbal Products, Thailand Institute of Scientific and Technological Research. The samples were analyzed using headspace GC-MS instrumentation (Agilent Technologies 7890 B), Triple Quad Mass Selective Detector 7000 D, capillary column set at HP-5 ms (30 m × 0.25 mm, film thickness 0.25 μm). The column temperature was set at 50°C - 230 °C, 4°C/min, injector mode pulse split 20:1, 230 °C, Detector MSD, EI 70 eV, scan mode, 40 – 400 amu, carrier gas was helium at 10.0 psi, flow 1.2 mL/min; an average velocity of 40 cm/sec was used for quantitative studies.

### Statistical analysis

Data are presented as mean ± SEM (standard error of mean). One-way ANOVA was employed for statistical analysis, followed by Dunnett’s test for multiple comparisons. Data were statistically significant at *p*-value <0.05.

### Ethical clearance

This study was approved by the Ethics Committee of Srinakharinwirot University under number SWUEC-246/2564F). All volunteers provided informed consent before participation.

## RESULTS

### Preference test of essential oil blend for stress with sleep problems

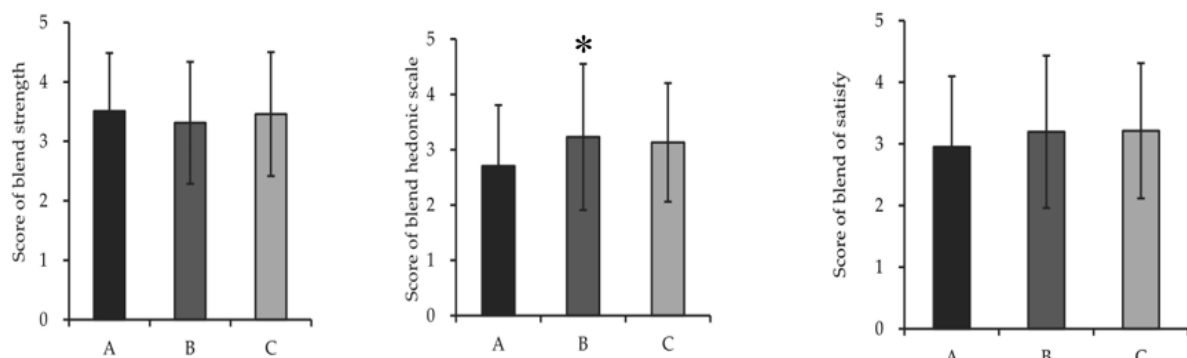
The majority of participants in each group (n=21 per group) reported the highest level of satisfaction with the essential oil blend Formula B, with a statistically significant difference compared to Formula A ( $p < 0.05$ ), which primarily contained Thai essential oils, including bergamot oil, peppermint oil, ylang-ylang oil, chamomile flower oil, and vetiver oil (Figure 1). Therefore, Formula B was developed further based on the volunteers' feedback, aiming to reduce the intensity of the scent and adjust the balance of the fragrance to make it more suitable for sleep and enhance relaxation.

The volunteer's opinion of formulation developments B1 and B2 is shown in Figure 2. There was no significant difference between B1 and B2, but B2 was selected due to its higher bergamot oil content to give freshness and unisex

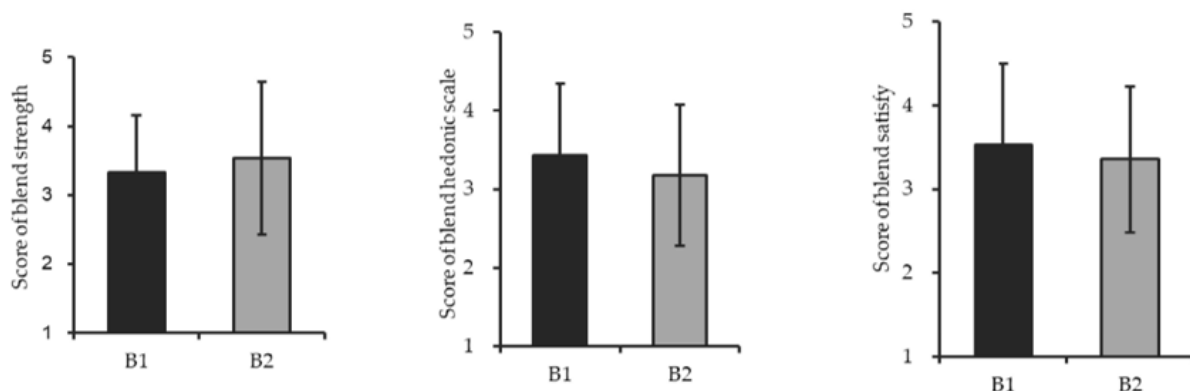
scent profile and reduce the floral middle note. The result of the blending was that B2 has a light citrus and freshness character compared with B1. Consequently, the B2 formulation may serve as a suitable candidate for future research aimed at examining the effects of essential oil blends on stress and sleep disturbances in greater detail.

### Aromatic chemical composition of essential oil blend using GC-MS

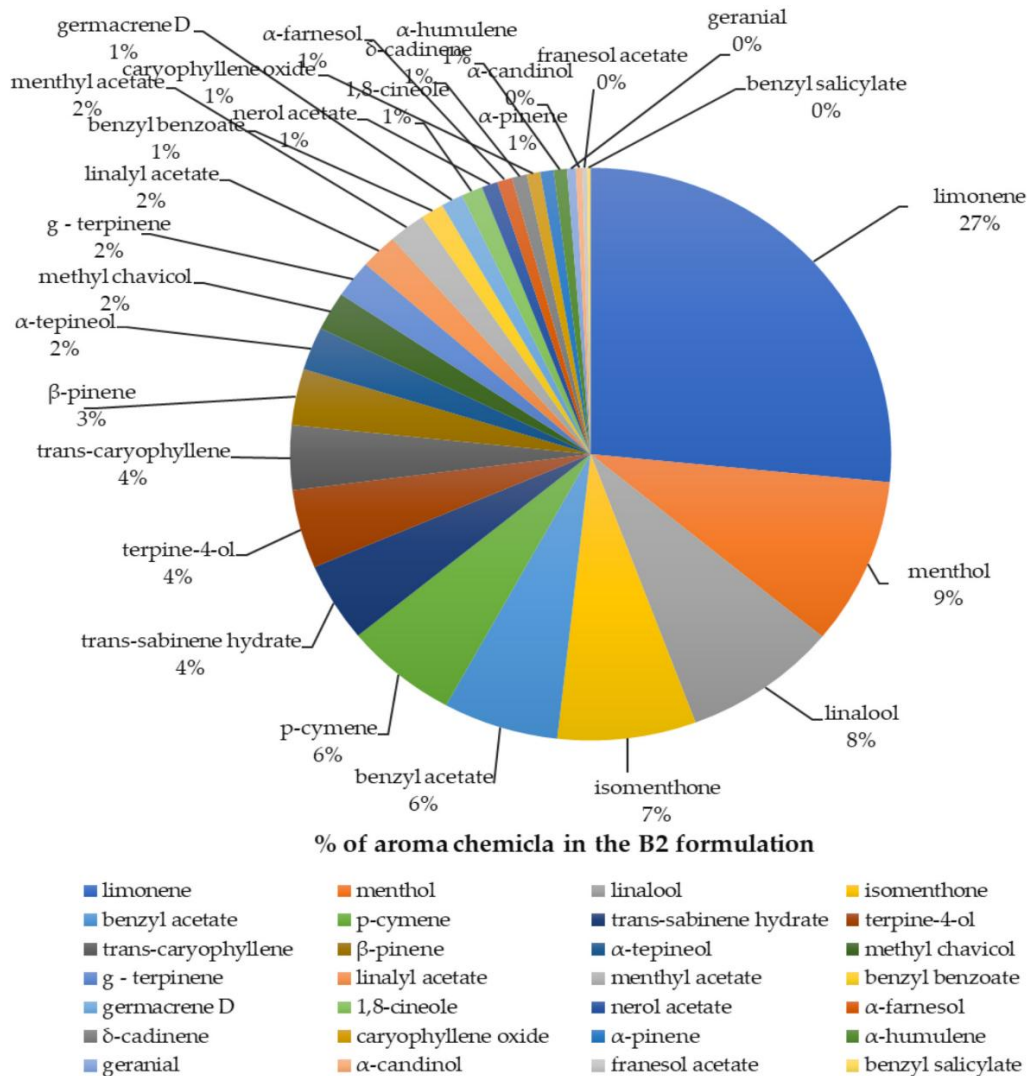
Essential oil samples were selected based on a preference test to assess the aromatic chemical composition of the blended oils (Figure 3). Table 4 shows the % peak area. Limonene, menthol, and linalool were the main components of the aroma in Formula B2. After improving the essential oil blend Formula B, volunteers preferred Formula B2 to B1, although no significant difference ( $p$  of blend strength = 0.820,  $p$  of hedonic scale = 0.309,  $p$  of overall satiation = 0.469) was noted among the preference evaluation of blend strength, hedonic scale, and overall satisfaction (Figure 2).



**Figure 1.** Preference test of essential oil blend for stress with sleep problem in volunteers. Three formulations (A, B, C) of essential oil blend were used. Expressed scores are on blend strength, blend hedonic scale, and blend satisfaction. Data expressed as mean  $\pm$  SEM (n=63) at  $p < 0.05$  (\*) was significant compared to A



**Figure 2.** Preference test of essential oil blend for stress with sleep problem in volunteers. Two formulations (B1 and B2) of essential oil blend were used. Expressed scores are on blend strength, blend hedonic scale, and blend satisfaction. Data are expressed as mean  $\pm$  SEM (n=63)



**Figure 3.** Ratio of aroma chemicals in essential oil blend formulation B2

## DISCUSSION

Although no significant difference was noted in the preference evaluation between B1 and B2, B2 was selected by the volunteers probably because it contained more bergamot oil, providing a fresh, unisex scent profile and reducing the intensity of the floral middle notes. GC-MS analysis revealed that the main components of B2 were limonene, menthol, and linalool. All these components have been reported to exhibit relaxation properties. Limonene, the principal component (26.56%), is known for its potential to reduce stress and improve mood disorders.<sup>(2,4)</sup> A previous animal study showed that the inhalation of navel orange *Citrus sinensis* (L.) Osbeck essential oil, which is rich in limonene, notably reduced depressive behaviors induced by chronic mild stress, lowered HPA axis activity, and

prevented a decline in monoamine neurotransmitters.<sup>(8)</sup> Additionally, it restored the downregulated expression of brain-derived neurotrophic factor in the hippocampus, indicating potential antidepressant effects of limonene.<sup>(8)</sup> Inhalation of clary sage (*Salvia sclarea* L.) oil has been shown to alleviate pain and promote relaxation in patients with periodontal disease<sup>(28)</sup> and reduce stress in female patients undergoing urodynamic testing.<sup>(29)</sup> These effects may be attributed to linalool and linalyl acetate, which are the key components of the oil. Menthol improves dopamine-mediated neurotransmission,<sup>(10)</sup> while various stress management interventions can lower blood cortisol levels and reduce anxiety.<sup>(30)</sup> Additionally, several studies have demonstrated that aromatherapy using linalool-rich essential oils, such as lavender (*Lavandula angustifolia* Mill.), bergamot (*Citrus bergamia*

Risso), and orange (*Citrus sinensis* L.), enhances mood and alleviates symptoms of stress, anxiety, and depression in patients.<sup>(31)</sup> Altogether, the essential oil blend, used in our study, was composed of aromatic chemicals that produce mood-enhancing effects due to the synergistic interaction of their properties.

The present study addressed the limitation that preference is inherently subjective and varies between individuals; therefore, determining the most suitable essential oil blend requires careful interpretation of personal responses to achieve an optimal balance. In this study, the selection of the essential oil blend was based on volunteer satisfaction through a preference test. The chosen formulation will be utilized in cosmetic product development and further investigated for its effects on additional physiological and psychological biomarkers in future experimental phases.

## CONCLUSION

In summary, the essential oil blend with a high concentration of bergamot oil was the top choice among volunteers, suggesting it may effectively support relaxation and sleep.

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## Author Contributions

Conceptualization, S.S., P.S. and K.S.; methodology, S.S., K.C., P.S., D.S., K.S.; software, K.S.; validation, P.S. and K.S.; formal analysis, S.S., R.R. and K.S.; data curation, K.S., P.S.; writing—original draft preparation, S.S., and K.S.; writing—review and editing, S.S., K.S. and P.S.; visualization, K.S. and P.S.; funding acquisition, S.S. and R.R. All authors have read and agreed to the published version of the manuscript.

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## Institutional Review Board Statement:

The study was conducted in accordance with the Declaration of Helsinki and approved by the Ethics Committee of Srinakharinwirot University (approval number SWUEC-246/2564F).

## Informed Consent Statement:

Informed consent was obtained from all subjects involved in the study.

## Data Availability Statement

Data is contained within the article and Supplementary Files.

## Conflicts of Interest:

The authors declare no conflicts of interest

## Declaration the Use of AI in Scientific Writing

No AI-assisted technologies were used in the writing, editing, or content generation of this manuscript. All content was produced solely by the authors.

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