



Evaluation of the awareness of the effects of aroma oils and assessment of the antioxidant and brightening effects

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Background: Aroma oils have various psychological and physical effects and have been used in the beauty industry in several ways. Although aroma oils have functional effects regarding skin beauty, they are not fully utilized in the cosmetic marketing field.

Objective: To bring out the potential of aroma oils in the market, the perception of efficacies of aroma oils and the evaluation of functional efficacy were studied and analyzed in this study.

Methods: The awareness of the effects of aroma oils was determined using surveys, and the actual cosmetic effects of aroma oils were analyzed via in vitro tests.

Results: As a result of evaluating the efficacy of aroma oils via experiments, basil, ylang–ylang, lemongrass, geranium, peppermint, and marjoram oils have antioxidant properties, and eucalyptus and lemongrass oils have brightening effects.

Conclusion: Basil, ylang–ylang, lemongrass, geranium, peppermint, and marjoram oils have the potential to be used as ingredients in anti–aging cosmetics. Also, eucalyptus and lemongrass oils have brightening effects, indicating that they have potential as ingredients for brightening cosmetics.

Keywords: 2,2–diphenyl–1–picrylhydrazyl assay; aroma oil; functional efficacy; perception; tyrosinase activity assay

Introduction

Aroma oils include essential oils extracted and processed from various parts of aromatic plants grown through natural or organic methods [1]. As these oils produce the plants' fragrances, they are found in small amounts in the cells of different parts of plants including the roots, flowers, stems, leaves, fruits, and peels. Therefore, the extraction of aroma oils requires a specific and complicated process [2,3], and their efficacy and properties differ depending on the extraction site. The chemical properties

of aroma oils continuously change in live plants [4,5], affecting the efficacy of the aroma oil. In the beauty industry, aroma oils are used for aromatherapy, a form of alternative medicine, as they are absorbed through the skin and the respiratory tract. Aroma oils are used as antioxidant and brightening agents and to treat acne and inflammation. They can also be used as the raw materials for functional cosmetics that meet the needs of domestic consumers.

The demand for functional cosmetics has grown steadily in the domestic cosmetics market. The production performance

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of functional cosmetics in the domestic market increased from 2.14 trillion won in 2012 to 4,443.9 billion won in 2016 (20.06%) [6]. This production increased by 2.5% in 2017 (4.85 trillion won) and 2018 (4.98 trillion won) [7]. In addition, the production of functional cosmetics with brightening effects increased by 62.90% from 2015 to 2016 (579.6 billion won) [6]. The production of anti-aging (antioxidant) cosmetics is expected to increase in the future as the country's population ages [8,9]. Therefore, aroma oils used as the raw materials for cosmetics have an unlimited potential in the domestic beauty industry and should be researched thoroughly.

Aroma oils are primarily used in aromatherapy due to their psychosomatic effects, and are also widely used as cosmetic materials to improve several skin problems such as rough skin and stress-related skin damage [4,5]. The chemical components of aroma oils differ depending on the extraction site, season, and time, which affect their absorption and mechanisms of [10,11]. Studies regarding the effects of aroma oils, the awareness of the effects of aroma oils, and the willingness of consumers to purchase aroma oils have been conducted [9], including a study regarding the perception and effects of aromatherapy using aroma oils [12] and another regarding the knowledge, perception, and utilization rate of aroma oils conducted to facilitate product research and development [1]. Based on these previous studies, this study attempted to determine the awareness of the effects of aroma oils to gauge the potential of their use in cosmetics.

Measuring the brightening effects of aroma oils

Melanin is the skin pigment that affects skin color and is formed by melanocytes [13]. Melanocytes have dendrites that extend to the keratinocytes distributed around them. Tyrosine is oxidized by tyrosinase within melanocytes to become dopa, which is oxidized to dopaquinone. Dopaquinone becomes eumelanin through leukodopachrome and dopachrome 5,6-dihydroxyindole [14-16]. Partial blockage of this process of melanin formation results in brightening of the skin. The tyrosinase activity inhibition method used in this study is an experimental evaluation method based on the principle that brightening can be achieved via the inhibition of tyrosinase in the melanin synthesis process. This method is widely used to evaluate the brightening effects [17,18], including in a study that evaluated a mixture of aroma oils (essential oils) in the inhibition of tyrosinase [19], and a study that examined the brightening effects of Ceylon cinnamon essential oils [20]. In this study, nine brightening effects of aroma oils were evaluated using the mushroom

tyrosinase inhibition assay.

Measuring the antioxidant effects of aroma oils

The inhibition of oxidation is referred to as antioxidizing. Cell aging is the same process as cell oxidation. Antioxidants are substances that delay or prevent oxidation (aging) [21]. The 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay is an experimental method used to evaluate antioxidant efficacy by measuring the degree of antioxidant activity using the degree of color change [22]. This method can be used to measure anti-aging activity that opposes free radicals as the measurement method is simple and closely related to the degree of antioxidant activity [23]. In this study, the DPPH assay was used to evaluate the antioxidant efficacy of aroma oils. This method was successfully used in a study regarding the antioxidizing efficacy of lemon grass oil [24] and a study regarding the antioxidizing activity of citrus essential oils [3].

Despite the unlimited potential of aroma oils in the cosmetic industry, they are not commonly used as raw materials in domestic cosmetics. Therefore, the brightening and antioxidant effects of aroma oils used in functional cosmetics were examined via an efficacy verification experiment and a survey regarding the perception of their efficacies. This study investigates and proposes ways to promote the use of aroma oils in functional cosmetics in the domestic market.

Materials and methods

Survey respondents and procedures

In this study, a survey was conducted to investigate the perception of the effects of aroma oils. The survey was conducted over a nine-day period in July 2020. A total of 427 questionnaires were included in the final analysis. The mushroom tyrosinase activity assay and DPPH assay were performed to verify the brightening and antioxidizing effects of aroma oils, respectively.

Data analysis method

Statistical analysis

The survey responses are presented as frequency and percentage and were analyzed using IBM SPSS statistical software, version 26.0 (IBM Corp., Armonk, NY, USA). Descriptive statistical analyses of the survey responses were also conducted. The credibility and validity of the questions included on the questionnaire were verified using Cronbach's alpha.

Mushroom tyrosinase activity assay

For the materials used for the experiment, 9 aromatic oils of eucalyptus, marjoram, geranium, lemongrass, tea tree, ylang-ylang, basil, peppermint, and clary sage were selected. They were selected because they are used a lot in domestic spas and esthetic shops and are familiar to consumers, and each oil is known to have the following effects.

Eucalyptus oil is good for cold, and strengthens immunity; marjoram oil is good for relaxation and warming; geranium oil is good for excretion of toxins, lymph circulation, and damaged capillaries; lemongrass oil works to promote digestion, relieve muscle pain and headache, and relieve stress; tea tree oil is effective for colds, sterilization, wounds and acne; ylang-ylang oil is effective for dry skin, high blood pressure, and menstrual pain; Basil oil is effective for headaches, varicose veins, and blood circulation.

To measure the inhibition of tyrosinase activity in eucalyptus, marjoram, geranium, lemongrass, tea tree, ylang-ylang, basil, peppermint, and clary sage, each aroma oil was dissolved in 99% ethanol to prepare 3.3%, 1.6%, 0.8%, and 0.4% aromatic oil solutions. A buffer mixture prepared using 0.1 M potassium phosphate buffer (pH=6.8) and L-tyrosine were added to the samples to reach a final volume of 150 μ l. After treatment with the assay reagent, the samples were incubated at 37°C for 30 minutes. Then, the absorbance at 490nm was measured using an ELISA reader.

DPPH assay

To measure the antioxidant activity of cedarwood, black pepper, geranium, and peppermint oils, 2.5%, 1.25%, 0.625%, and 0.313% aromatic oil solutions were prepared using 99% ethanol (99%). Basil, thyme, cypress, lemongrass, clary sage, tea tree, lavender, marjoram, and ylang-ylang oils were prepared at concentrations of 1.25%, 0.625%, 0.313%, and 0.156%, based on their solubility in ethanol. Each sample contained 10 μ l of aromatic oil solution and 190 μ l DPPH (1 M). The samples were incubated at 37°C for 15 minutes. Then, the absorbance at 517 nm was measured using X. The free radical scavenging efficacy of DPPH was expressed as a percentage of the difference in absorbance between the control group and the experimental group divided by the absorbance of the control group.

Results and Discussion

Respondent characteristics

Of the 427 respondents, over one-third (35.4%; 151 respon-

Table 1. Respondent demographics

Characteristic	Number (total=427)	Percentage (%)
Age (yr)		
20-29	99	23.2
30-39	98	23.0
40-49	79	18.5
\geq 50	151	35.4
Skin type		
Normal	63	14.8
Dry	112	26.2
Oily	78	18.3
Combined problems	143	33.5
Sensitive	31	7.3
Occupation		
Student	47	13.3
Office worker	128	36.3
Researcher	18	5.1
Professional	81	22.9
Other	79	22.4
Monthly income (won)		
<2 million	30	7.0
2-3 million	67	15.7
>3-4 million	81	19.0
>4-5 million	56	13.1
>5 million	193	45.2
Total	427	100.0

dents) were aged 50 years or older while 23.2% (n=99) were 20-29 years old, 23.0% (n=98) were 30-39 years old, and 18.5% (n=79) were 40-49 years old (Table 1). Normal skin was reported by 14.8% (n=63) of respondents. Combined skin problems were most commonly reported by respondents (33.5%; 143 respondents), followed by dry skin (26.2%; 112 respondents), oily skin (18.3%; 78 respondents), and sensitive skin (7.3%; 31 respondents).

Awareness of the brightening effects of aroma oils

The majority of respondents (73.2%) were not aware of the brightening effects of aroma oils. Respondents aged 20-29 years were most aware of the brightening effects (37.4%), followed by respondents aged 50 years or more (29.1%). Most students (51.1%) were aware of the brightening effects, as were nearly one-third of respondents with an income <3 million won (31.6%). Previous studies have reported that the brightening effects of aroma oils are excellent [5]. As most survey respondents were unaware of the brightening effects of aroma oils, marketing regarding these effects should be increased.

Awareness of the antioxidant effects of aroma oils

Nearly half of the respondents (45.0%) reported awareness of the antioxidant effects of aroma oils. The awareness of the antioxidant effects was the highest among respondents aged 50 years or more (52.3%) and the lowest among those aged 30–39 years (33.0%). Nearly half of respondents with sensitive skin (51.6%), professionals (50.5%), respondents with combined skin problems (50.3%), and students (48.9%) were aware of the antioxidant effects. While the awareness of the antioxidant effects of aroma oils was found to be higher than that of the brightening effects of aroma oils, it was low. Antioxidant effects have been reported for several aroma oils [25]. More specific marketing regarding the antioxidant effects is necessary.

Awareness of the effects of aroma oils

Over half of the respondents (51.1%) reported awareness of the moisturizing effects of aroma oils while 28.3% reported awareness regarding the antioxidant effects (Table 2). The moisturizing effects of aroma oils were the most recognized effects by respondents regardless of age, occupation, income level, or skin type. A previous study reported that 33.3% of consumers reported that the most prominent effects of aroma oils leading to their purchase were anti-aging effects and 23.9% of consum-

ers cited moisturizing effects as the cause of their purchase of aroma oils [9], reflecting the consumer's desire to appear youthful. These data are correlated with the aging population [26] and suggest the need for the development of anti-aging products.

Assessment of the anti-oxidant effects of aroma oils

Basil, geranium, peppermint, marjoram, ylang-ylang, and lemongrass aroma oils were found to have antioxidant effects via the inhibition of free radical activity. The mean DPPH antioxidant scavenging activity of basil oil was 83.0% ($p < 0.001$) in a 0.16% basil oil solution, 90.0% ($p < 0.001$) in a 0.31% basil oil solution, 92.0% ($p < 0.001$) in a 0.63% basil oil solution, and 94.0% ($p < 0.001$) in a 1.25% basil oil solution (Fig. 1). These are consistent with the antioxidant effects of basil oil reported in a previous study [27]. Basil aroma oils are rich in phenolic compounds and contain various polyphenols, including anthocyanins, and natural substances, including flavonoids [27].

The mean DPPH antioxidant scavenging activity of ylang-ylang oil was 56.0% ($p < 0.001$) in 0.16% ylang-ylang oil solution, 70.0% ($p < 0.001$) in a 0.31% ylang-ylang oil solution, 87.0% ($p < 0.001$) in a 0.63% ylang-ylang oil solution, and 96.0% ($p < 0.001$) in a 1.25% ylang-ylang oil solution (Fig. 2). Ylang-ylang oil contains monoterpene, sesquiterpenes, and phenyl-

Table 2. Reported awareness of aroma oil effects

Division	Brightening	Antiaging	Moisturizing	Acne treatment	Sensitivity treatment	χ^2 (p)
Age (yr)						44.225*** (<0.001)
20–29	11 (11.1)	19 (19.2)	46 (46.5)	10 (10.1)	13 (13.1)	
30–39	1 (1.0)	25 (25.8)	59 (60.8)	1 (1.0)	11 (11.3)	
40–49	9 (11.4)	22 (27.8)	36 (45.6)	2 (2.5)	10 (12.7)	
≥50	10 (6.6)	54 (35.8)	80 (53.0)	1 (0.7)	6 (4.0)	
Occupation						17.310 (0.138)
Student	9 (19.1)	10 (21.3)	22 (46.8)	3 (6.4)	3 (6.4)	
Office worker	7 (5.5)	42 (32.8)	64 (50.0)	5 (3.9)	10 (7.8)	
Professional	4 (4.0)	26 (26.3)	55 (55.6)	2 (2.0)	12 (12.1)	
Other	6 (7.6)	21 (26.6)	39 (49.4)	4 (5.1)	9 (11.4)	
Monthly income (won)						12.931 (0.114)
<3 million	6 (6.1)	28 (28.6)	47 (48.0)	7 (7.1)	10 (10.2)	
3–5 million	11 (8.1)	32 (23.5)	71 (52.2)	5 (3.7)	17 (12.5)	
>5 million	14 (7.3)	61 (31.6)	103 (53.4)	2 (1.0)	13 (6.7)	
Skin type						21.241 (0.169)
Normal	0 (0.0)	14 (22.2)	42 (66.7)	2 (3.2)	5 (7.9)	
Dry	9 (8.0)	38 (33.9)	53 (47.3)	2 (1.8)	10 (8.9)	
Oily	12 (15.4)	22 (28.2)	34 (43.6)	3 (3.8)	7 (9.0)	
Combination	7 (4.9)	38 (26.6)	77 (53.8)	6 (4.2)	15 (10.5)	
Sensitive	3 (9.7)	9 (29.0)	15 (48.4)	1 (3.2)	3 (9.7)	
Total	31 (7.3)	121 (28.3)	221 (51.8)	14 (3.3)	40 (9.4)	-

Values are presented as n (%).

*** $p < 0.001$.

propanoids and has antioxidant effects [11].

The mean DPPH antioxidant scavenging activity of lemongrass oil was 27.0% ($p < 0.001$) in a 0.16% lemongrass oil solution, 37.0% ($p < 0.001$) in a 0.31% lemongrass oil solution, 52.0% ($p < 0.001$) in a 0.63% lemongrass oil solution, and 75.0% ($p < 0.001$) in a 1.25% lemongrass oil solution (Fig. 3). Lemongrass oil has antioxidant properties, and contains 75%–85% citral (3, 7-dimethyl-2, 6-octadien-1-al), z-citral, borneol, estragole, methyleugenol, geranyl acetate, geraniol, beta-myrcene, limonenepiperitone, citronellal, citrate-2, alpha-terpineole, pinene, farnesol, proximadiol, and (+)-cymbodiactal [28].

The mean DPPH antioxidant scavenging activity of geranium oil was 10.0% ($p < 0.01$) in a 0.31% geranium oil solution, 14.0% ($p < 0.01$) in a 0.63% geranium oil solution, 21.0% ($p < 0.001$) in a 1.25% geranium oil solution, and 38.0% ($p < 0.01$) in a 2.50%

geranium oil solution (Fig. 4). In a previous study, geranium oil had superior antioxidant scavenging effects when compared to ascorbic acid [29]. Geranium oil contains citronellol (29.90%), trans-geraniol (18.03%), 10-epi- γ -eudesmol (8.27%), isomethone (5.44%), and linalool (5.13%).

The mean DPPH antioxidant scavenging activity of peppermint oil was 11.0% ($p < 0.001$) in a 0.31% peppermint oil solution, 15.0% ($p < 0.001$) in a 0.63% peppermint oil solution, 22.0% ($p < 0.001$) in a 1.25% peppermint oil solution, and 33.0% ($p < 0.001$) in a 2.50% peppermint oil solution (Fig. 5). The antioxidant activities of domestic peppermint oils and international peppermint oils have been reported as 5.7% and 97.8%, respectively [30].

The mean DPPH antioxidant scavenging activity of marjoram

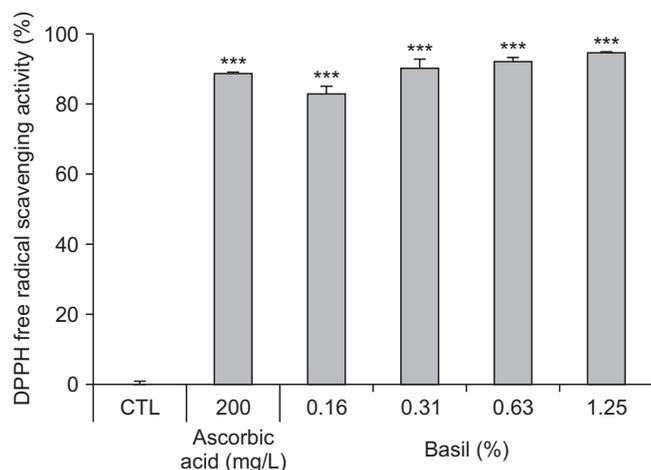


Fig. 1. Antioxidant effects of basil oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. ***Statistically significant ($p < 0.001$).

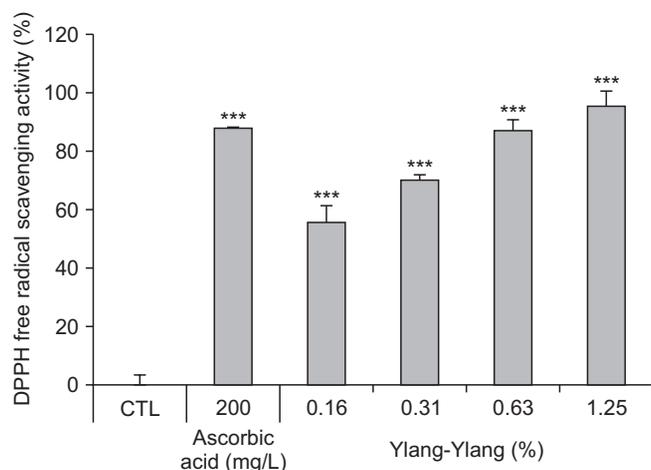


Fig. 2. Antioxidant effects of ylang-ylang oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. ***Statistically significant ($p < 0.001$).

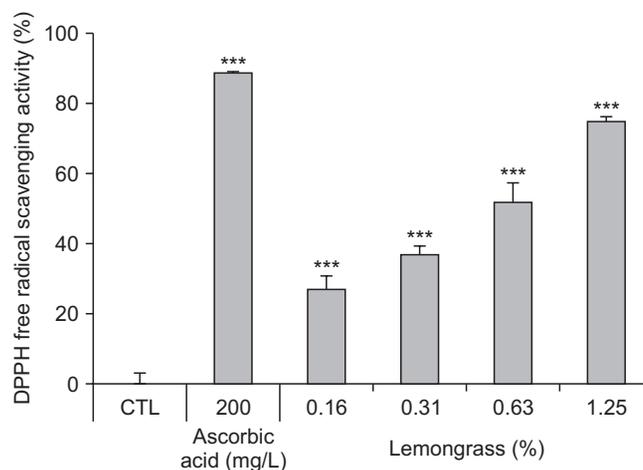


Fig. 3. Antioxidant effects of lemongrass oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. ***Statistically significant ($p < 0.001$).

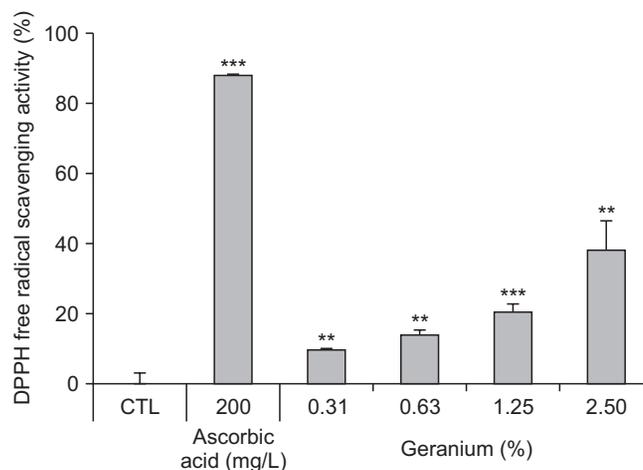


Fig. 4. Antioxidant effects of geranium oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. Statistically significant (** $p < 0.01$, *** $p < 0.001$).

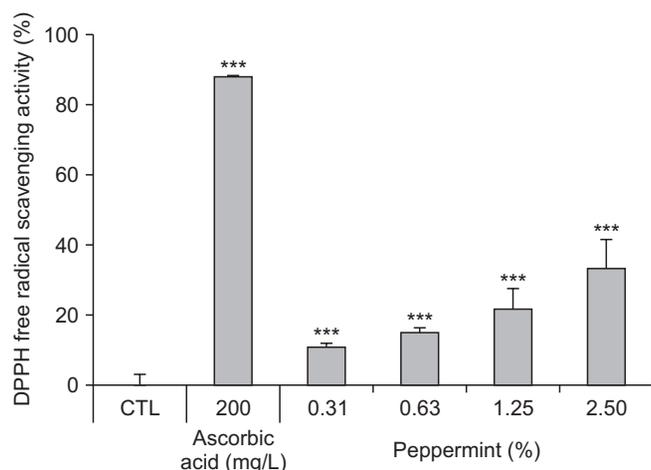


Fig. 5. Antioxidant effects of peppermint oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. ***Statistically significant ($p < 0.001$).

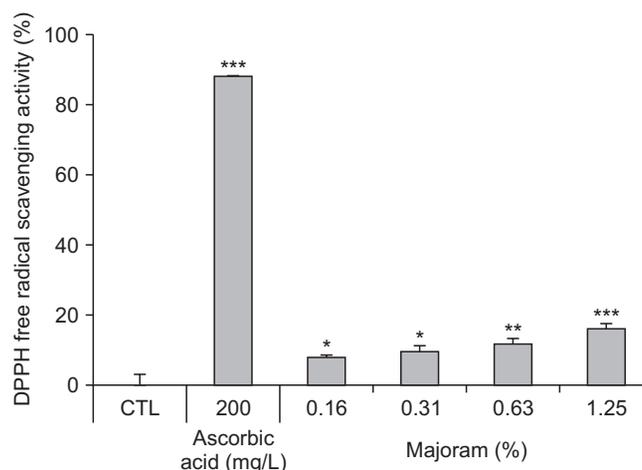


Fig. 6. Antioxidant effects of marjoram oil. DPPH, 2,2-diphenyl-1-picrylhydrazyl; CTL, control. Statistically significant (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

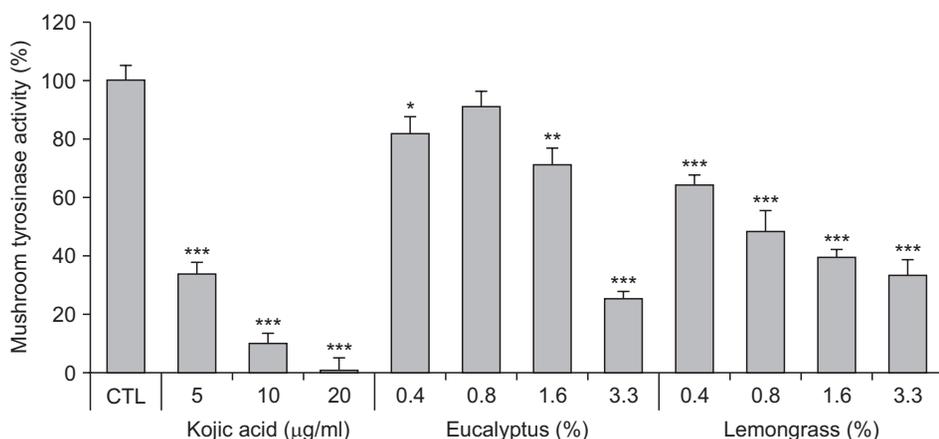


Fig. 7. Brightening effects of eucalyptus and lemongrass oils. CTL, control. Statistically significant (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

oil was 8.0% ($p < 0.05$) in a 0.16% marjoram oil solution, 9.0% ($p < 0.05$) in a 0.31% marjoram oil solution, 12.0% ($p < 0.01$) in a 0.63% marjoram oil solution, and 16.0% ($p < 0.001$) in a 1.25% marjoram oil solution (Fig. 6). Marjoram contains unstable antioxidants including carnosic acid, carnosol, RA, CA, flavonoids, luteolin-7-O-glucoside, and apigenin-7-O-glucoside [31]. The chemical components of marjoram oil include terpinen-4-ol, γ -terpene, and α -terpineol, which have antioxidant properties [31].

Assessment of the brightening effects of aroma oils

Lemongrass and eucalyptus aroma oils inhibited tyrosinase activity, which results in brightening of the skin in vivo. The brightening effects of lemongrass oil increased in a concentration-dependent manner, as the melanin production was 34.0% ($p < 0.001$) in a 3.3% lemongrass oil solution, 40.0% ($p < 0.001$) in a 1.6% lemongrass oil solution, 48.0% ($p < 0.001$) in a 0.8% lem-

ongrass oil solution, and 64.0% ($p = X$) in a 0.4% lemongrass oil solution (Fig. 7). Lemongrass oil contains limonene, which has lightening and brightening properties [32].

The melanin production was 26.0% ($p < 0.001$) in a 3.3% eucalyptus oil solution, 71.0% ($p < 0.01$) in a 1.6% eucalyptus oil solution, 91.0% in a 0.8% eucalyptus oil solution, and 82.0% ($p < 0.05$) in a 0.4% eucalyptus oil solution (Fig. 7). As the melanin production is significantly different between the 3.3% and 1.6% eucalyptus oil solutions, a 3.3% eucalyptus oil solution should be used as a brightening agent. The chemical composition of eucalyptus oil is 34.9% oxygenated monoterpenes, 31.8% oxygenated sesquiterpenes, 29.0% monoterpene hydrocarbons, and 4.3% sesquiterpenes hydrocarbons. Eucalyptus oil inhibits melanin synthesis, indicating that it should be included in skin care products [33].

This study analyzed the awareness of the effects of aroma oils and assessed the antioxidant and brightening effects of aroma

oils. The results of this study indicate that additional marketing regarding the effects of aroma oils is necessary in the cosmetic industry and that aroma oils should be included in the development of skin care products. Most respondents in this study were not aware of the brightening and antioxidant effects of aroma oils, suggesting that these effects should be better advertised. The most well-known effects of aroma oils were their moisturizing and antioxidant effects, though the awareness of these effects was low. Therefore, these properties can be used in consumer marketing or product development to promote the use of aroma oils. Through this study, the aromatic oils used in the experiment were proved to have antioxidant effects in the order of basil, ylang-ylang, lemongrass, geranium, peppermint, and marjoram oils, and after eucalyptus, lemongrass oil was proven to have a whitening effect. Like this as aroma oils have antioxidant and brightening effects, they should be included in skin care products. Aroma oils are traditionally used as alternative medicines that affect the nervous system. When they are included in cosmetic products, the user may also experience these effects.

This study is not without limitations. As the respondents in this study were most familiar with the moisturizing and antioxidant effects of aroma oils, an assessment of the moisturizing effects should be conducted. Only domestic consumers were included as respondents in this study. An international survey should be conducted due to the increased use of aroma oils worldwide.

Conflicts of interest

The authors have nothing to disclose.

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