ผลของน้ำมันหอมระเหยบางชนิดที่ใช้มากในประเทศไทย ต่อสรีรวิทยา และอารมณ์ความรู้สึก

นายวินัย สยอวรรณ

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรดุษฎีบัณฑิต
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EFFECTS OF SELECTED VOLATILE OILS COMMONLY USED IN THAILAND ON PHYSIOLOGICAL ACTIVITIES AND EMOTIONS

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A Thesis Submitted in Partial Fulfillment of the Requirements

for the Degree of Doctor of Philosophy Program in Public Health Sciences

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Thesis Title	EFFECTS OF SELECTED VOLATILE OILS COMMONLY USED IN THAILAND ON PHYSIOLOGICAL ACTIVITIES AND EMOTIONS
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วินัย สยอวรรณ: ผลของน้ำมันหอมระเหยบางชนิดที่ใช้มากในประเทศไทยต่อสรีรวิทยาและอารมณ์ ความรู้สึก. (Effects of Selected Volatile Oils Commonly Used in Thailand on Physiological Activities and Emotions) อ. ที่ปรึกษาวิทยานิพนธ์หลัก: รศ. ดร. นิจศิริ เรืองรังษี, อ. ที่ปรึกษาวิทยานิพนธ์ร่วม: รศ. ดร.นัยพินิจ คชภักดี, รศ. ดร.ฐาปนีย์ หงส์รัตนาวรกิจ, 132 หน้า.

ปัจจุบัน มีการใช้น้ำมันหอมระเหย อย่างแพร่หลายในประเทศไทยแต่ขาดผลพิสูจน์ในเชิงวิทยาศาสตร์ วัตถุประสงค์ของการศึกษาครั้งนี้ เพื่อทดสอบผลของน้ำมันหอมระเหยที่ใช้มากในประเทศไทย ได้แก่ น้ำมันลาเวนเดอร์ น้ำมันโรสแมรี่ น้ำมันมะลิและน้ำมันตะไคร้หอม ต่อระบบประสาท ได้แก่ ประสาทส่วนกลาง ประสาทส่วนอัตโนมัติรวมทั้งการตอบสนองของอารมณ์หลังจากการสูดดม อาสาสมัครจำนวน 20 คน ได้รับน้ำมันหอมระเหยหนึ่งกลิ่น ดังนั้นรวมอาสาสมัครทั้งหมด 80 คน พารามิเตอร์ระบบประสาทอัตโนมัติที่ทดสอบ ได้แก่ ความดันโลหิต การเต้นของหัวใจ อัตราการหายใจ และอุณหภูมิที่ผิวหนัง สำหรับระบบประสาทส่วนกลางมีการศึกษาการเปลี่ยนของคลื่นสมองโดยบันทึกข้อมูลทั้งความถี่ ค่าฟูเรียร์ทรานส์ฟอร์มอย่างเร็ว และแผนภาพคลื่นสมอง นอกจากนั้นการตอบสนองทางอารมณ์ได้ถูกประเมินโดย visual analog scale เปรียบเทียบผลการเปลี่ยนแปลงของระบบประสาท และอารมณ์ความรู้สึกระหว่าง น้ำมันหอมระเหย และน้ำมันอัลมอนด์โดยใช้สถิติ paired t-test และวิเคราะห์ความสัมพันธ์ระหว่าง ระบบประสาท กับอารมณ์ความรู้สึกโดยใช้ สถิติ Spearman rank correlation ผลการศึกษาพบว่ากลิ่นลาเวนเดอร์และกลิ่นตะไคร้หอม ทำให้การทำงานของระบบประสาทอัตโนมัติลดลง กลิ่นลาเวนเดอร์ทำให้ คลื่นสมองประเภท ธีต้า แอลฟ่าเพิ่มขึ้น ในขณะที่กลิ่นตะไคร้หอมทำให้คลื่นสมองทั้งแอลฟาและเบต้า เพิ่มขึ้น ในทางตรงกันข้าม กลิ่นโรสแมรี่ และกลิ่นมะลิกระตุ้นการทำงานของระบบประสาทอัตโนมัติเพิ่มขึ้น กลิ่นโรสแมรี่ทำให้คลื่นสมองประเภท แอลฟ่าลดลง แต่เบต้าเพิ่มขึ้น แต่กลิ่นมะลิทำให้ คลื่นสมองชนิดเบต้าเพิ่มขึ้น ในส่วนของอารมณ์ความรู้สึกพบว่า อาสาสมัครรู้สึกว่าตนเองมีความรู้สึกดีมากขึ้น หลังจากได้รับน้ำมันหอมระเหยทุกชนิด นอกจากนี้ อาสาสมัคร ยังรู้สึกกระตือรือร้น กระปรี้กระเปร่า และผ่อนคลายมากกว่าเมื่อเปรียบเทียบกับการดมน้ำมันอัลมอนด์ ในด้านความสัมพันธ์ระหว่างการตอบสนองทางอารมณ์กับระบบประสาทอัตโนมัติพบว่าอารมณ์สดชื่นมีความสัมพันธ์เชิงบวกกับการ ทำงาน ของระบบประสาทอัตโนมัติ ในทางตรงข้ามกัน อารมณ์ รู้สึกดี สงบนิ่ง ง่วงซึมมีความสัมพันธ์เชิงล บกับการทำงานของระบบประสาทอัตโนมัติ ความสัมพันธ์ระหว่างการตอบสนองทางอารมณ์กับคลื่นสมอง พบว่ามีทั้งความสัมพันธ์เชิงบวก (ความผ่อนคลายกับคลื่นสมองแบบแอลฟา) และเชิงลบ (คลื่นสมองแบบเบต้ากับความผ่อนคลาย) สำหรับความรู้สึกสดชื่น มีความมีความสัมพันธ์ในทางกลับกัน ผลการทดลองครั้งนี้สามารถใช้เป็นข้อมูลสนับสนุนเชิงวิทยาศาสตร์สนับสนุนว่าน้ำมันหอมระเหยมีผลต่อมนุษย์ทั้งในด้านร่างกายและอารมณ์ความรู้สึก

สาขาวิชา วิทยาศาสตร์สาธารณสุข ลายมือชื่อนิสิต.....

ปีการศึกษา 2554 ลายมือชื่อ อ.ที่ปรึกษาวิทยานิพนธ์หลัก..... .....

ลายมือชื่อ อ.ที่ปรึกษาวิทยานิพนธ์ร่วม.....

ลายมือชื่อ อ.ที่ปรึกษาวิทยานิพนธ์ร่วม.....

5279408153 : MAJOR PUBLIC HEALTH SCIENCES

KEYWORDS : ESSENTIAL OIL, BRAIN WAVE, EEG, AUTONOMIC NERVOUS SYSTEM

WINAI SAYORWAN: EFFECTS OF SELECTED VOLATILE OILS COMMONLY USED IN THAILAND ON PHYSIOLOGICAL ACTIVITIES AND EMOTIONS. ADVISOR : ASSOC. PROF. NIJSIRI RUANGRUNGSI, Ph.D., CO-ADVISOR ASSOC. PROF. NAIPHINICH KOTCHABHAKDI., Ph.D., ASSOC. PROF. TAPANEE HONGRATANAWORAKIT, Dr.rer.nat., 132 pp.

Nowadays, Volatile oils have been widely used in Thailand without much supporting scientific evidence. The objective of this study is was to investigate the effects of the commonly use volatile oil in Thailand, such as lavender oil, rosemary oil, jasmine oil and citronella oil on the nervous system, i.e. central nervous system (CNS), autonomic nervous system (ANS) as well as on emotional response after inhalation. Twenty subjects were tested for each essential oil. Totally eighty subjects were participated in this study. ANS parameters, i.e. blood pressure, heart rate, respiratory rate, and skin temperature were recorded. CNS was monitored by recording of brain electrical activities. Data were collected including frequency, Fast Fourier Transform value and topographical mapping. In addition, emotional responses were evaluated by visual analog scales. The effects of oils on the nervous system and emotional responses were determined by comparing the mean values between the oil and sweet almond oil. The paired t- test was used in this study. Correlation analyses between the nervous system and emotional responses were performed by Spearman rank-order correlation coefficient. Results demonstrate that lavender odor and citronella odor decreased the function of ANS. Lavender odor increased theta and alpha waves whereas citronella odor increased both alpha and beta waves. In contrast, rosemary odor and jasmine odor activated the function of ANS. Rosemary odor decreased alpha level and increased beta wave whereas jasmine odor increased beta wave. For emotional responses, subjects felt very good after inhalation all odors. The oils caused significant increases of enthusiasm, freshness and relaxation, when compared with sweet almond oil. Correlation between emotional responses and ANS showed a positive correlation between freshness and the increase of ANS function. In contrast, emotion of good, calm, drowsy had a negative correlation with ANS function. The correlation between emotional responses and brain wave showed both a positive correlation (relaxation and alpha brain wave) and a negative correlation (beta brain wave and relaxation). In terms of freshness, the correlation has been observed in opposite direction. Results from this study are able to be scientific knowledge of the effects of volatile oil on human body and emotion.

Field of Study : Public Health Sciences

Academic Year : 2011

Student's Signature .....

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LIST OF ABBREVIATIONS

µg	Microgram
µl	Microliter
ANS	Autonomic nervous system
ANOVA	Analysis of variance
BR	Breathing rate
BP	Blood Pressure
BMI	Body Mass Index
°C	Degree Celsius
Ca^{2+}	Calcium ion
Cen	Center
cm	Centimeter
Cl^-	Chloride ion
CO	Citronella oil
DC	Direct Current
Dias BP	Diastolic Blood Pressure
EEG	Electroencephalograpy, Electroencephalogram
EC	Eye Closed
EO	Eye open
FFT	Fast Fourier Transformation
g	Gram
GC /MS	Gas chromatography / Mass spectrometer
GMOS	The Geneva Emotion and Odor Scale
HEOG	Horizontal electro-oculo-gram
hrs	Hours
IPSP	Inhibitory Postsynaptic Potential

JO	Jasmine oil
LA	Left anterior
LO	Lavender oil
LP	Left posterior
m^2	Square meter
mg	Milligram
min	Minute
MEG	Magnetoencephalography
ml	Milliliter
mm	Millimeter
msec	millisecond
mV	millivolt
Na^+	Sodium ion
R	Rest
RA	Right anterior
RO	Rosemary oil
RP	Right posterior
SD	Standard derivation
Sys BP	Systolic Blood Pressure
SO	Sweet almond oil (Base oil)
VAS	Visual analog scale
VEOG	Vertical electro-oculo-gram

CHAPTER I

INTRODUCTION

Background and Significance of the Study

An essential oil is a concentrated volatile aromatic compound derived from plants. Owing to the difference in environmental conditions and neighboring fauna and flora, each plant species nurtured in a certain country has specific characteristics. Essential oil can be extracted from oil 'sacs' in flowers, leaves, stems, roots, seeds, wood and bark [1- 2]. It is widely known that the odor of essential oils can be used to treat illnesses, namely a therapy referred to aromatherapy. Aromatherapy can then be defined as a therapy that uses aromas. More accurately, aromatherapy is a branch of botanical medicine using volatile and aromatic plants compounds that has been considered as a treatment tool for various conditions. It has been thought that essential oils have certain effects on the person inhaling it [2- 3]. In Thailand, the result from the study of Patin demonstrated that the most popular essential oils used for aromatherapy by inhalation and massage included lavender (35.71%), rosemary (28.2%), citronella (20.54%) and jasmine (12.50%) respectively [4]. Furthermore, it has been shown by the Division of Complementary and Alternative Medicine at the Ministry of Public Health that lavender (88.6 %), jasmine (63.6%), citronella (56.8%) and rosemary (50%) were the main essential oils sold in Bangkok [5].

Smell is an important sensory perception in mammals. Based on the knowledge in physiology, the olfactory perception originating in nasal cavities is divided into three main parts of nostril, respiratory segment, and olfactory segment. In general, the olfactory system perceives odorant molecules via specialized sensory cells. In order to transmit odorant information throughout the olfactory bulb, an enlarged segment of the first cranial nerve (CN I), the odorant molecules then expose to the olfactory segment and later trigger the olfactory bipolar neurons. Subsequently, the olfactory bulb transmits odorant information via olfactory tract to the olfactory tubercle that are connected to several areas of the brain such as the perform cortex, amygdale, and lateral entorhinal cortex. Presumably, these afferent pathways could be involved with several emotion and physiological effects [6- 7].

Evaluation of the aroma effects on the nervous system can be divided into two different forms of arousal; the central nervous system (brain wave activity) and the autonomic arousal (heart rate, blood pressure, skin temperature, respiratory rate, etc.) [8]. The measuring the neurological activity of brain structures is through the use of electroencephalography (EEG). This difference of brain activity response to specific odors for example EEG showed that alpha waves, which are associated with relaxation, increased in the presence of relaxing odors such as lavender. Moreover, it has been reported that the frontal alpha waves reduced during the state of alertness [9-11]. EEG results indicated that olfactory stimulation affected to the physiological response of the central nervous system. As a result of the peripheral nervous system function, it has been observed that autonomic central nervous system changes in response to aversive olfactory stimulation. Measurement by an alteration of the autonomic arousal is interpreted in terms of a sedative/relaxing effect of aromas [8]. The researcher found that the sympathetic and parasympathetic nervous systems were affected by odorant stimulation, as measured by the electrocardiogram. This is further supported by reports of a peripheral vassal constriction as a result of olfactory stimulation. Blood pressure has also been noted as being reduced by relaxing scents such as lavender and ylang-ylang oil. In contrast, the increases of the cortical arousal and/or the autonomic arousal are interpreted in terms of a stimulating effect of aromas. Rosemary and jasmine for example, significantly increased breathing rate as well as systolic and diastolic blood pressure [12-15].

Smells in the environment have many effects on the psychological well-being, moods and behaviors. Positive emotions have been observed as the increase of juxtaposition activities in the entorhinal cortex (a limbic structure) but negative emotions linked with activation of the medial thalamus and left orbital frontal cortex. Generally, emotional study in the laboratory conditions have been shown that smelling of pleasant odors resulted in an increase of bilateral activities in the occipitotemporoparietal cortex, lateral cerebellum, hypothalamus, anterior temporal cortex, amygdala, and hippocampus [16]. It has also been reported that essential oil, such as rosemary, activated alertness but lavender odorant was an anti-stress agent and reduced the arousal state. Moreover, there were uses of lavender and orange oil to

decrease level of anxiety and improve mood in dental clinic, after insomnia women inhale lavender oil significantly improvement in sleep quality [17-20].

Many researchers studied the effect of volatile oil on brain wave activity, the autonomic nervous system and emotions. However, these findings were contradictory, for example, Diego and his colleagues found that individuals felt more relaxed and an improved emotion after inhaling lavender oil. Moreover, an increase of mid frontal (F3, F4) alpha power on their EEG was found after inhalation of the oil. Motomura suggests that lavender has been demonstrated to decrease stress scores and increase Theta 1 (3.5-5.5 Hz) brain wave activity but decrease Beta 1 (13.5-20 Hz) which is associated with relaxation [9]. In contrast, Masago found that there was a partial decrease in alpha 1 (8-11 Hz) activity and a significant decrease in posterior temporal lobe activity after receiving lavender oil [21]. Some researches about autonomic nervous system activity also showed contrasting results. For example, Tongnit and her colleague found a significantly decreased blood pressure, heart rate and respiratory rate caused by 3 minutes inhalation of lavender essential oils [12]. However Sriboon reported that inhalation lavender oil by aroma lamp caused a significant decrease in respiratory rate and subjective calmness, relaxation, but diastolic blood pressure and heart rate increased [22]. Rosemary and jasmine affected to autonomic nervous system in administration of transdermal form [14-15]. There was no experiment about effect of rosemary and jasmine on autonomic nervous systems parameter and emotional response after inhalation technique was been reported. There are studies supported that the mode of administration could make a significant difference in the resulting effect for example; East Indian Sandalwood; whose main constituent is α-santalol. The oil induced alertness when applied via inhalation, yet it reduced physiological arousal when massaged via transdermal [23]. Furthermore, citronella has not been formerly reported about its effects on brain waves. In order to gain more reliable results, this study was designed to strictly control hedonic effect (pleasant and unpleasant) which was reported earlier that hedonic effect had an influence on physiological effect [16]. In the research by Brauchli *et al.*, they reported that heart rate is an autonomic variable which can be affected by pleasant and unpleasant oils. For example, valeric acid (judged unpleasant) increase heart rate, but heart rate decreased with phenylethyl alcohol (rated pleasant) in order to control the hedonic

effect [24]. Before the experiment, they were asked to inhale base oil and lavender oil to rate the pleasantness of the smell on a five-point Likert scale. The participants, who indicated oil pleasantness within the target level range of 2 -4 were chosen to participate in the study.

There are only a few recent studies in Thailand about the effects of essential oil on physiological and emotional activities. In addition, transdermal techniques have been used in most experimental researches to convey essential oil (e.g. rosemary, orange oil, and ylang-ylang oil, jasmine oil) into the body [13-15, 25]. From reviewing previous literature demonstrates that this study seems to be the first experimental research in Thailand to examine physiological effects of essential oils on autonomic nervous system and emotions by inhalation and on central nervous system by EEG. Thus, the purpose of this study is to determine effects of essential oil in three dimensions on; the central nervous system (brain wave), the autonomic nervous system (heart rate, blood pressure, breathing rate and skin temperature) and emotions.

Research questions

1. Whether essential oil has effects on central nervous system?
    1.1. Whether essential oil affects to brain wave frequency?
    1.2. Whether essential oil affects to brain wave amplitude?
2. Whether essential oil has effects on autonomic nervous system?
    2.1. Whether essential oil affects to heart rate?
    2.2. Whether essential oil affects to blood pressure?
    2.3. Whether essential oil affects to skin temperature?
    2.4. Whether essential oil affects to breathing rate?
3. Whether essential oil has effects on emotions?
4. Whether essential oil could affect differently to central nervous system, autonomic nervous system and emotions?

Hypothesis

Based on the review of previous researches, it could be hypothesized that

1. Lavender and citronella induce relaxant activities as following:
    1.1. Autonomic nervous system could decrease heart rate, blood pressure and breathing rate. But it may increases skin temperature.
    1.2. Alpha wave activity could be increased.
2. Rosemary and jasmine induce alertness activities leading to:
    2.1. Autonomic nervous system could increase heart rate, blood pressure and breathing rate. On the other hand, it may decrease skin temperature.
    2.2. Beta wave activity could be increased.

Objectives of the Study

1. To investigate the changes of electroencephalogram in subjects after inhalation of essential oil.
2. To investigate the changes of autonomic nervous system in subjects after inhalation of essential oil.
3. To investigate the changes of emotion in subjects after inhalation of essential oil.

Expected Benefits

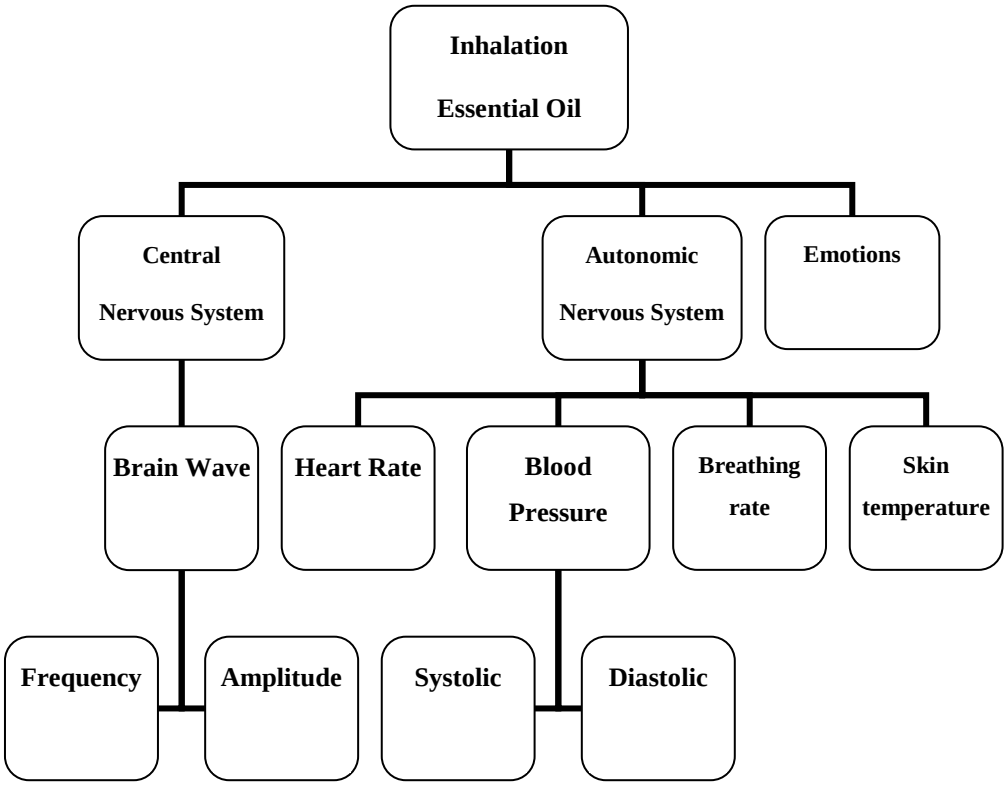
1. This study will be useful for selecting the most appropriate essential oil for certain out comes. For example, the best essential oil for fleshing condition can increase alpha wave and heart rate. Moreover, the best essential oil for relaxation must decrease heart rate but increase beta waves. Finally, cognitive conditions can be induced by the essential oil that increases theta wave.
2. To provide information related to safety and awareness of essential oils that increase blood pressure for people who have clinical history about

high blood pressure. Furthermore, drivers should avoid essential oils inducing relaxing and sleepy in the car.

3. This protocol could be applied for further researches in order to study effects of other essential oils and their mixture on physiological effects and emotions.

Conceptual Frameworks

The conceptual frameworks of this research are scoped in a chart as shown below;



CHAPTER II

LITERATURE REVIEWS

The literature review of this study was described in five main parts as the followings;

- Human olfactory systems
  - Anatomy and physiology of human olfactory system
  - Variation of the olfaction perception
  - Test of olfactory function
- Nervous systems
  - Central nervous system and Electroencephalography (EEG)
  - Autonomic nervous system and Autonomic nervous system measurements
- Emotional state
- Volatile oils properties
- Related reviews

Human olfactory system

Anatomy and physiology of human olfactory system

Any study on the perception of odors by human must begin with an examination of the anatomy and physiology of the human olfactory system, as shown in figure 1 on the location and structure of the olfactory receptors. Receptors for smell (olfaction) are specialized to respond to volatile chemicals that have been dissolved in the mucous coating of the nasal cavity. The olfactory receptors contained within a region of the nasal mucosa called the olfactory epithelium that is located in the upper portion of the cavity on side of the nasal septum. Tiny hairs (cilia) are the receptive portions of the olfactory receptor cells. It is believed that these cilia provide several different receptor sites for odorous molecules to interact with the cell. This interaction causes the cell to be depolarized and generate a general potential that causes a nerve impulse to be conveyed directing to the brain via the olfactory nerve. This nerve

impulse triggers a response from two areas of the brain; the thalamus and the limbic systems. The thalamus initiates a conscious perception and a fine discrimination of an odor through the cortex. However, the exact basis for this discrimination is largely unknown. The limbic system coordinates certain behavioral and emotional responses to particular odors. These responses to the odorous stimuli vary significantly from person to person. Due to of the natural complex nature of the anatomy and physiology involved, odor perception is very subjective and difficult to standardize and measure quantitatively [7, 26-28].

It is generally assumed that olfactory receptor sites are on the ciliary surface membrane. Odorant stimuli bind to a protein receptor site in the membrane. The stimulus activated receptor activates G-proteins which evoke an enzyme cascade. At the end channel proteins are phosphorylated that may affect gating of ion channels. Until now specific receptors have been found for a number of odor qualities. It is summed that about 100 to 300 receptor classes exit and each cell is more and less sensitive to each odorant and, therefore, a great variety of combinations are possible. It is said that the human being can differentiate about 1000 odors with differing qualities. To date, it is not possible to predict an odor sensation due to the chemical structure of an odorant with a view to establishing an odorant classification system. The axons of the receptor cells form bundles, called olfactory nerves or olfactory filaments. This arrangement allows the synchronous excitation of a number of cells, which are not close neighbors. This enhances stimuli of lower intensity. Lateral inhibition procesess at subsequent cell layers suppress intense and long lasting signals. This phenomenon is called peripheral adaptation, which protects humans from stimulus overflow [27-30].

The filaments enter the olfactory bulbs where they synapse with the dendrites of mitral cells. Several hundreds of primary olfactory axons converge on a single mitral cell. The information is already processed here. From the bulbs olfactorius the second and their order neurons pass by the limbic system and the thalamus to the projection area of the brain. Feedback loops of efferent nerves allow modification of the stimuli, which causes central adaptation [29-31].

The perception of odors is a pre-condition of odor annoyance. The annoyance reaction of an exposed person however is also determined by non-sensory variables such as personality traits, attitude to the source, environmental context, etc.

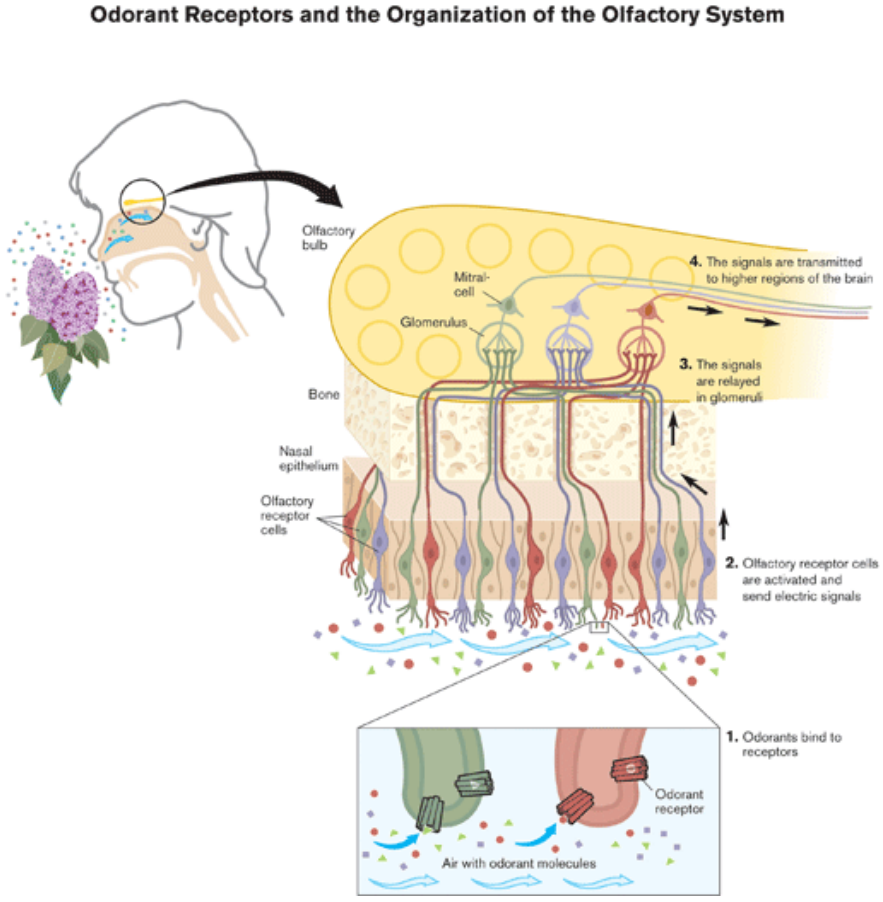


Figure 1 Location and structure of the olfactory receptors [32].

Variation of the olfactive perception

Sex

In most of the studies reviewed in this report, sex differences were not considered, so an analysis of this issue is limited. However, in an extended replication of the test, both orange and lavender essential oil in contrast to a music or no-odor condition, and found that both odors were able to improve better mood and reduce anxiety equally among men and women. It is unclear why this inconsistency in sex differences occurred or what it means; the authors themselves do not offer any explanation [33-34]. Goel *et al.* also found differences in the way that men and

women's sleep was affected by the scents of lavender and peppermint, respectively [35]. However, here too, the effects could not be predicted across experiments. It is most likely that when sex differences are observed, they are due to an interaction between psychological and physiological factors. At this time the basis for these differences has not been well elucidated. The degree to which one is susceptible to the emotional connotation of an odor and forming associations to it will modulate the effectiveness of an aroma to influence mood, physiology and behavior. This researcher found that women are more sensitive than men to odors at certain times during the menstrual cycle, and this varying sensitivity may modulate the effectiveness of aromas on physical and emotional states [36]. In this study select equal amount of male and female, and exclude women who presently have menstruating.

Age

An age-related decline in olfactory function has been demonstrated by a number of psychophysical measures including odor detection threshold magnitude estimation odor discrimination, odor identification and odor recognition memory.

Anatomical changes associated with aging have been detected in both peripheral olfactory structures, e.g., olfactory bulb, but their relative contributions to the age-related functional deficits are unknown. The effects of age have been examined in one study which showed a decline in amplitude with increasing age. The researcher showed that normal adults from 20 to 50 years of age could identify 85-100% of the odor presented. Determination of odor identification began about age 50 and 60 years old adults could identify for 65-70% of these odors. From 60 years old, begins a decline of the sensation, the discrimination and the identification of the smells. More of half of the persons with more than 80 years old have a bad smell among which 25% of the subjected smell nothing more [37-38]. In this study, selected subjects were aged between 18-35 years.

Hormone

The olfactive perception is influenced for a woman, by sexual hormones. It is increased during the ovulation and at the beginning of the pregnancy. On the other hand, a decline of its small is observed in the course of menstruation, at the end of pregnancy, and after the menopause. However, Evelia Navarrete-Palacios *et al.* conducted that threshold differed significantly across the cycle and was lowest during the ovulatory and highest during the menstrual phase. Odor detection thresholds were determined using sniff bottles containing –log 9.5 to –log 6.0 concentrations of amyl acetate presented in ascending order. Thresholds differed significantly across the cycle and were lowest during the ovulatory and highest during the menstrual phase. Thresholds for all control groups were higher than those for the cycling women during the ovulatory phase [39-40]. The results confirm that olfactory threshold is related to phase of the menstrual cycle and thus possibly to hormonal state.

Obesity

Andrzej Obrebowski concluded that in 30 children, aged 10-16 years and suffering from simple obesity, significantly lowered odor detection thresholds were note. The thresholds were lower than the average for a given age group in around 20% of obese children in cases of odors stimulating olfactory nerve and in around 57% in cases of substances stimulating olfactory and trigeminal nerves [41]. Odor identification test was similarly affected, with identification of olfactory nerve plus trigeminal nerve stimulating odors affected more than twice as frequently. The detected alterations may be linked to metabolic disturbances, which accompany with simple obesity [42]. Thus, this experiment was done by selecting subjects with BMI between 18-25 kg/m^2.

Health of human

The perception may change through diseases (e.g. cold), toxic damages of the olfactory cells (e.g. through drugs) or forced impact on the skull (head), usually it deteriorates. Permanent impacts of odor substances on the olfactory cells lead to a deterioration of the sensitivity on account of adaptation. These processes are described as an adaptation of habituation. Some persons smell, for example, an odor

in th e p lace o f another (dysosmia) o r s till id entify a s mell w hich d oes n ot e xist (phantosmia). The most painful confusion stays the loss of smell (anosmia). The most frequent causes result f rom a cr anial t raumatism w ith d estruction o f th e o lfactive nerve, t he na sal i nfections (chronic r hinitis) or f rom a di sease of A lzheimer dur ing which the "reading" of the smells by the brain becomes impossible [43-45]. Thus, this experiment was selected for healthy subjects.

Smoking tobacco/ coffee and tea

The non smokers are more sensitive than the smokers which concern about the respiratory s ystem. C affeine (in coffee) an d t heophylline (in t ea) had influence on increasing s ynaptic functions, resulting in the decrease threshold of the nerve ending cell [46-47].

Variation of the ol factive perception is information for s creening s ession. Inclusion criteria s ubjects aged be tween 18 a nd 35 years w ith normal body m ass indices. N one o f s ubjects h ad ab normalities af fecting s mell, car diovascular d iseases, or a history of smoking or drug addiction. Subjects were screened for a n ormal sense of smell u sing th e n-butyl a lcohol te st method and w ho w ere m enstruating w ere not included i n the study. The d ay b efore ex periment, researcher w ill co ntact t he participants by phone to confirm the experiment date. Before the experiment starting, subjects m ust s hampoo t heir ow n ha ir. A pplication of ha ir s pray, a ntiperspirants or perfumes was not a llowed. A dditionally, pa rticipants s hould a void alcohol, caffeinated and tea drinks.

Test of olfactory function

Olfactory testing h as been o ften neglected in a clinical p ractice despite t hat it may of fer va luable information i n t he otolaryngologic a nd ne urologic clinical examination [45]. T his m ay be a ttributed to the l ack of s imple, f ast, a nd r eliable methods of ol factory evaluation. T o fulfill t his need, s everal olfactory t ests w ere introduced dur ing t he pa st t wo decades, t hese s everal t ests ar e w idely u sed in clinically and were chosen including;

University of Pennsylvania Smell Identification Test [UPSIT; known commercially as the Smell Identification Test™ was developed at smell test in the USA center and can be self-administered in 10 to 15 minutes by most patients in the waiting room and scored in less than a minute by nonmedical personnel. This 40-item test, along with its briefer clones, is available in numerous languages and has been employed in hundreds of clinical and experimental studies. In this test, a patient was presented with 40 "scratch and sniff" odorant pads and is required to choose, from four response alternatives, an answer for each stimulus, even if none seems appropriate or no odor is perceived. Olfactory function can also be classified, on an absolute basis, into six categories: normosmia, mild microsmia, moderate microsmia, severe microsmia, anosmia, and probable malingering [48].

Threshold olfactory tests typically employ a dilution series of a stimulus in an odorless diluents, such as

Single Ascending Series Butanol Odor Threshold Test. The stimuli used in this standardized test consist of 12 ternary aqueous dilution steps of *n*-butanol (from a 4% v/v initial dilution mixture) presented in ascending order in a two-alternative forced-choice paradigm. The threshold was defined as the lowest concentration at which a subject correctly indicated which of two plastic squeeze bottles-one containing odorant and other the diluents-produced the stronger odor on five consecutive trials [49-50].

Phenyl ethyl alcohol single-staircase odor detection threshold test. In this test, detection threshold values for the rose-like odorant phenyl ethyl alcohol were determined by using a modified single-staircase procedure as described in detail elsewhere. In the present study, the staircase was begun at the –6.5log concentration step of a half-log step (v/v) dilution series extending from –10.00log concentration to –2.00log concentration. It was moved upward in full –log steps until correct detection occurred on five sets of consecutive trials at a given concentration. If an incorrect 6 response was given on any trial, the staircase was moved unward a full-log step. When a correct response was made on all five trials, the stair-case was reversed and subsequently moved up or down in half –log increments or decrements, depending upon the subject's performance on two pairs of trials (each pair consisting of a choice between a blank and an odorant) at each concentration step. The geometric mean of

the first four staircase reversal points following the third staircase reversal was used as the threshold measurement[49, 51].

Odor discrimination test. In this test, a subject was presented with 16 sets of three microencapsulated odorant (two same, one different) on separate pages of a cardboard test booklet. The stimuli on a given page of the test were presented in rapid succession and the examinee was asked to select the “odd” or “different” odor within each trial. The odorants of a triad were preselected to be equivalent in average perceived intensity, as determined from intensity ratings presented elsewhere. The number of triads in which the different stimulus was correctly reported served a dependent measure [52].

In this study, we used single ascending series butanol odor detection threshold test for selecting subjects before the test. Subjects can be distinguished two odors (n- butyl alcohol and water) at concentration that lower than Step 6(5.48×10^{-3} v/v) of n- butyl alcohol in water.

Nervous system

Central nervous system

The central nervous system (CNS) plays a critical role in a short and long-term regulation of arterial pressure. The short-term regulation is exerted from second to second by modulation of an activity of sympathetic postganglionic and cardiovagal neurons. The long-term arterial pressure regulation is also exerted by hormonal and neural control of blood volume. The medulla oblongata is the central integrative area in controlling the circulation. The role of neurons at precisely defined regions are generating the tonic excitatory background transmitted to spinal preganglionic and maintaining normal resting levels of arterial pressure, integrating most reflexes involved in arterial pressure regulation, coupling signals generated in higher brain during various behavior to optimize circulatory pattern, sensing metabolic and hormonal signal for regulating arterial pressure and serving as the main target of drugs acting to lower arterial pressure. The neurons in the rostral ventrolateral medulla (RVLM) are parasympathetic neurons which may be the major sensory area

of excitatory input to vasomotor neurons. Their basal discharge rate related to mean arterial blood pressure and vasomotor tone. The resting discharge is continually modified by a variety of peripheral inputs.

Nucleus tractus solitaries (NTS) in the meduall oblongata is the site of the first synapse of the afferent baroreceptor fibers which form part of the 9^{th} and 10^{th} cranial nerves. Apart from the baroreceptors, somatic & nucleus afferents arterial chemoreceptor, cardiopulmonary receptors and inputs from hypothalamus and sensor in motor cortex also synapse in the NTS. Although afferent inputs are widely dispersed through many polysynaptic pathways to numerous central integrative areas, the NTS projects directly to the spinal cord, parabrachial nucleus, nucleus ambiguous and hypothalamus. The vasomotor center receives inhibitory input from the NTS in contrast to the excitatory fivers which innervate the dorsal motor nucleus of the vagus. Thus, an increase activity of baroreceptor due to elevated blood pressure leads to a decrease activity in the sympathetic efferent systems and increase vagal tone. Higher centers involved in cardiovascular regulation include areas of the hypothalamus, basal ganglia, limbic system, and cerebral motor cortex. Stimulation of the posterior hypothalamus elevates blood pressure which is mediated through an increase in the tonic activity of the vasomotor center [53-55].

Electroencephalography (EEG)

Source of EEG Activity and Principle Mechanism

The EEG recorded at any region of the scalp approximately quantifies the total activity of pyramidal neurons in a certain volume of tissue under the electrode (Figure 2). Pyramidal neurons are the major projection neurons in the cortical cortex.

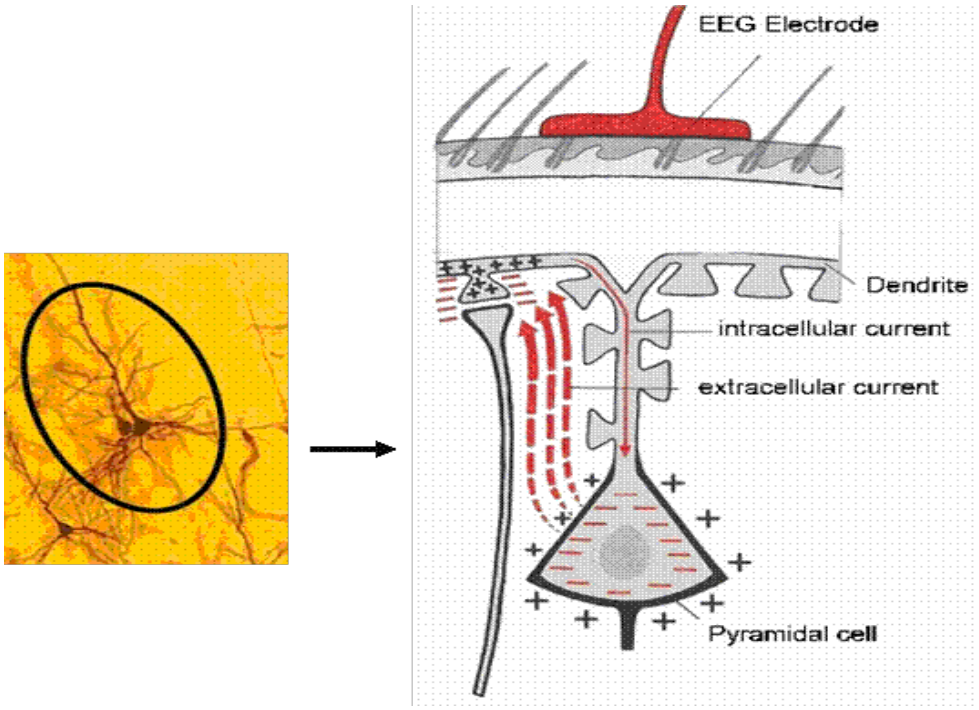


Figure 2 The diagram illustrates a pyramidal cell which is the major source of electrical current which is found in EEG data [56].

Their apical dendrites are oriented perpendicular to the cell body and receive a variety of synapse inputs. The ion exchange in the pyramidal neurons is the principle source of EEG activity. Cation (Sodium (Na^+) potassium (K^+)) and anion (large intracellular ions e.g., Chloride (Cl^-), protein) relate to the generation of electrical brain activity. During resting state, the inside of the cell provides the membrane resting potential of -65 mV. Most K^+ is inside the cell but the Na^+ concentration outside the cell is higher than inside. When the neuron is excited to reach the threshold that causes a gate in the Na^+ channel to open. Due to the high concentration of Na+ outside, Na^+ diffuses into the neuron. The electrical potential changes to about +40 mV. The membrane resting potential is reduced that, leading to the depolarization at the cell membrane. The Na^+ channels become refractory and K^+ channels open. When they do open, K^+ rushes out of the cell, reversing the depolarization. Also at about this time, sodium channels start to close. This causes the action potential to go back ward to -60 mV. This action is later followed by repolarization as K^+ flows into the cell and Na^+ is pushed outward. The membrane potential will return to the resting state again [56-57]as shown in Figure 3.

The propagation of the action potential reaches to the postsynaptic membrane and leads to the depolarization of the postsynaptic cell, it is called excitatory postsynaptic potential (EPSP). If that signal leads to the hyperpolarization of the postsynaptic cell, it is called inhibitory postsynaptic potential (IPSP). Both types of synapses make contact with a neuron. The summation of potential at postsynaptic site will be either depolarization or hyperpolarization. If the depolarization reaches a threshold level, an action potential will be launched. The EEG signal probably comes from postsynaptic cells. The electrical activity of the EEG requires thousands of neurons to be synchronously activated to generate a signal large enough to be measured by EEG. However, not all cells contribute equally to the EEG because it predominantly reflects the activity of cortical neurons that close to the surface of the skull.But the deep structures (e.g., hippocampus, thalamus, brain stem) do not contribute directly to the surface [58-59].

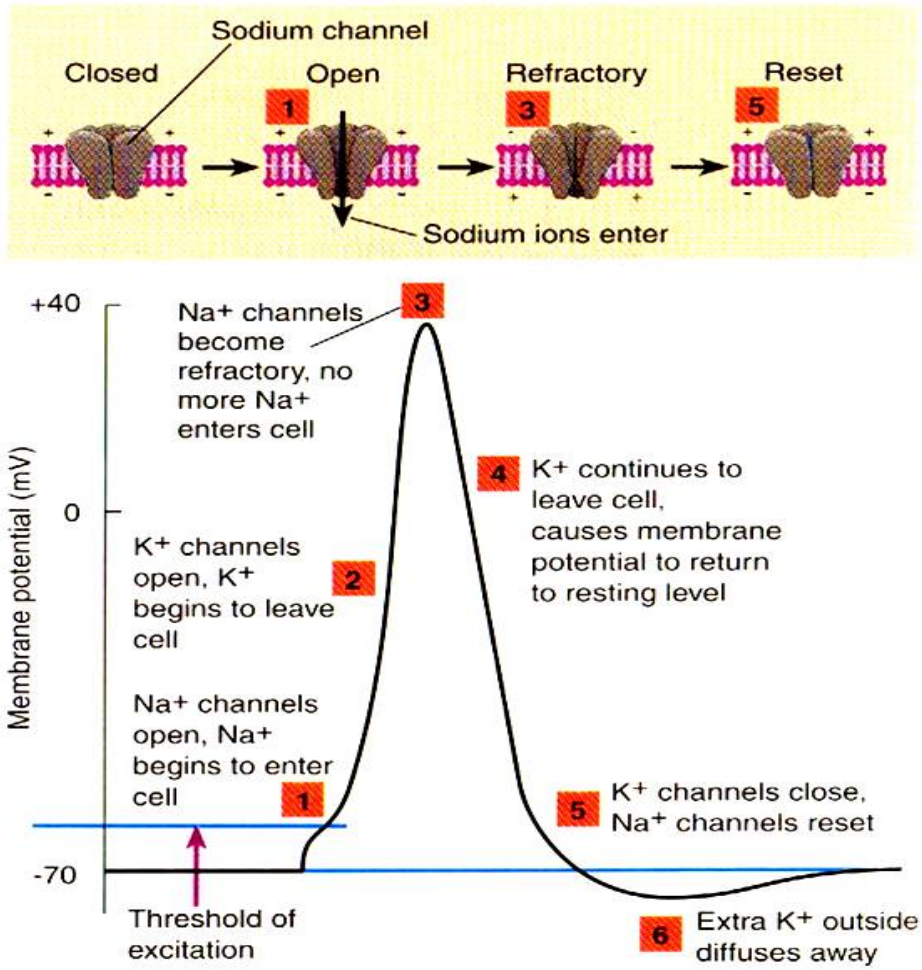


Figure 3 Step of action potential 1. At rest outside of the membrane is more positive than inside. 2. Na^{+} moves inside the cell causing an action potential, the influx of positive sodium ions makes inside of membrane more positive than the outside. 3. Potassium ions flow out of the cell, restoring the resting potential net charges. 4. Sodium ions are pumped out of the cell and potassium ions are pumped into the cell, restoring the original distribution of ions[58].

General basic of the EEG signal

The EEG signal comes from the large neural population in the cerebral cortex synchronized together to summate at the scalp surface. However, recordable voltage at the scalp surface is reduced by the meninges, cerebro-spinal fluid, skull and scalp tissue,and is measured in microvolts (μV). The signal describes voltage differences between electrodes and a reference electrode on the skull over a certain time range. The neuroelectrical signal will be transformed from an analogue to a digital signal to display the biological signal in the computer. However, the neuro-electrical signal to very small and needs to be amplified. The signal gets further adjusted to get clear, by means of gain, sensitivity and filtering. The EEG signal is described in its frequency (Hz, cycles per second) and amplitude (μV, microvolt).

The amplitude of the EEG signals changes depending on spatial (position) and temporal (time) parameters as shown in figure 4. The EEG amplitudes of the scalp EEG lie between 10-100 μV, and about 1-2 mV (millivolt) when measured on the surface of the brain. Moreover, the size of amplitude depends on how synchronous is the activity of neurons under that brain area. The more neurons are excited simultaneously, the greater the increase in amplitude. While EEG provides a rather poor spatial resolution (because of the limited number of electrodes and the distortion of the signal after passing through the volume conductor such as bone and brain tissue), it gives very high temporal resolution (in milliseconds). It thus is greatly sensitive to even minor change of brain activity. The following EEG-related-terms, which are usually referred to, are shortly described [58-61].

Frequency: It refers to the temporal dimension of EEG activity (Hertz; Hz).

Voltage (volt): It refers to the potential difference between two locations.

Morphology: It refers to the shape of the waveform. The shape of a wave or an EEG pattern is determined by the frequencies that combine to make up the waveform and by their phase and voltage relationships.

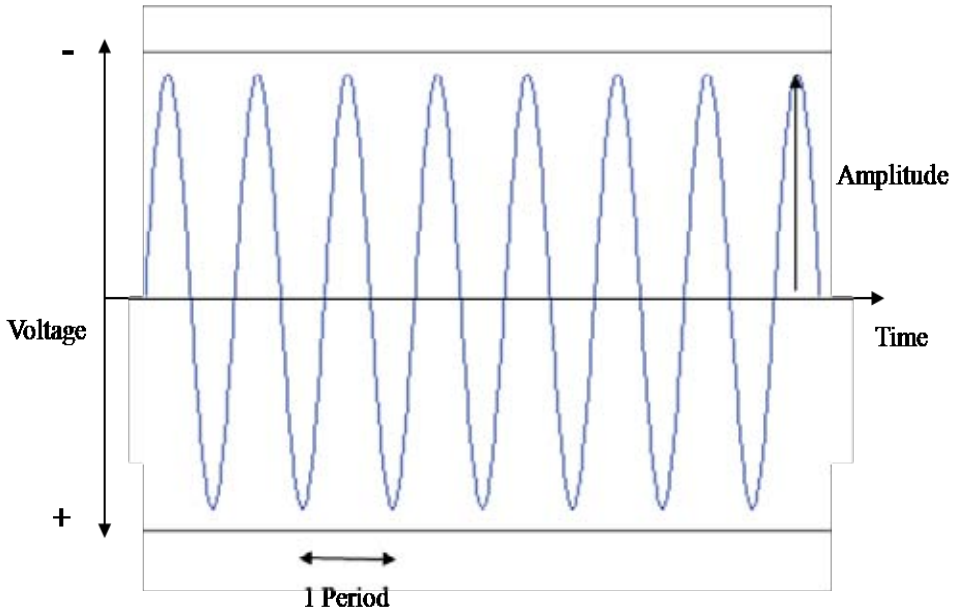


Figure 4 The EEG amplitude and frequency

The EEG signal is described as the frequency measured in Hertz (Hz) and amplitude which can divide brain wave into four types [58-59, 61].

Alpha: rhythm at 8-12.9 Hz occurs during wakefulness over the posterior regions of the head, generally with higher voltage over the occipital area. Amplitude is extremely variable and generally under 50 microvolts in adults. It has been seen with closed eyes and under conditions of the physical relaxation and the relative mental activity.

Beta: brain wave has the highest frequency between 13 and 30 Hz. This is associated with our normal waking state. Beta helps in logical thinking, analysis and active attention function.

Theta: rhythm at 4-7.9 Hz, associated with intuition, is known as the 'sixth sense' which allows people to access their subconscious. It is activated during dream sleep and a deep meditation state. Theta is also associated with creative thinking, recalls, intuition and allows people to tap into their inner genius.

Delta: has the lowest frequency between 0.5 and 3.9 Hz. Delta is produced during deep sleep.

Autonomic Nervous system

The autonomic nervous system is a part of the nervous system that controls the visceral functions of the body. This system helps to control arterial blood pressure, gastrointestinal motility and secretion, urinary output, sweating, body temperature and many other activities, some of which are controlled almost entirely by the autonomic nervous system and some only partially controlled. The autonomic nervous system is composed of two divisions: sympathetic and parasympathetic [53-54, 62].

The sympathetic nervous system

Preganglionic cells of the sympathetic division extend from the first thoracic segment to lower lumbar segments of the spinal cord (thorocolumbar outflow). The cell body of the preganglionic neurons are found within the intermediolateral column of spinal cord and axons of preganglionic neurons emerge from the spinal cord to the paravertebral chain ganglia. A preganlionic fiber may synapse with many postganglionic neurons that are often distributed among different paravertebral ganglia. This divergence permits coordinated activation of sympathetic neurons at several spinal levels. The axons of postganglionic neurons within the paravertebral ganglia travel in the spinal peripheral nerves to autonomic targets. Some neurons of the cervical and upper thoracic ganglia innervate peripheral vessels, sweat glands, and hair follicles, while others innervate visceral organs and glands of head and chest as well as heart, lung, and vascular smooth muscle. Lower thoracic and lumbar paravertebral ganglia innervate peripheral blood vessel, sweat gland, and pilomotor smooth muscle. Some preganlionic fibers pass the paravertebral sympathetic ganglia and branches of the splanchnic nerves to synapse on neurons of the prevertebral ganglia. The prevertebral sympathetic ganglia innervate the gastrointestinal system and the accessory gastrointestinal organs, including kidneys, pancreas, liver, and also provide the major sympathetic innervations of the bladder and external genitalia. Another group of preganglionic fibers run with the spanchnic nerve into the abdomen and innervates cells of the adrenal medulla, which are developmentally and functionally related to postganglionic sympathetic neurons [53-54].

The parasympathetic nervous system

The parasympathetic preganglionic neurons are located within the brain stem and in the S_2–S_4 segments of the spinal cord (cranio-sacral outflow). Their axons are longer than those of postganlionic neurons. The parasympathetic preganglionic neurons within both the brain stem and spinal cord project to postganglionic neurons in ganglia which reside close to visceral targets. This is contrast with sympathetic postganglionic neurons within the para-or prevertebral ganglia which are distant from their targets. Parasympathetic preganglionic fibers, such as the vagus, innervate parasympathetic ganglionic neurons, which in turn innervate the target, tissues. The axons of the motor neurons in the dorsal vagus nucleus project in the vagus nerve to postganglionic neurons in thoracic and abdominal targets, such as lungs, esophagus, stomach, livers, gall bladder, pancreas, and upper intestinal tract. Neurons of the ventrolateral nucleus ambiguous provide the principal parasympathetic innervations of the cardiac ganglion, which innervates the heart. The axons of parasympathetic preganglionic cell bodies in the sacral spinal cord project to the pelvic ganglion plexus. Pelvic ganglion neurons innervate the descending colon, bladder, and external genitalia [53-54].

The effects of sympathetic and parasympathetic innervations of each organ are relatively well-established, and are summarized in Table 1.

Table 1 Examples of the effects of sympathetic or parasympathetic stimulation on various organs [54].

Organ	**Sympathetic**	**Parasympathetic**
Heart	Increase heart rate	Decrease heart rate
Blood vessels	Constriction	Dilatation
Lung Bronchi	Relaxed	Constricted
Stomach		
Motility	Decrease	Increase
Sphincter	Increase tone	Relaxed
Secretion	Inhibition	Stimulation
Eye		
Pupil	Dilated	Constricted
Intestine	Motility reduced	Digestion increased
Bladder	Sphincter closed	Sphincter relaxed

Autonomic nervous system measurements

Heart rate (HR)

Heart rate is a non-invasive assessment of cardiac autonomic nervous system. It refers to beat-to-beat alterations in heart rate which is defined by the degree of balance in sympathetic and vagus nerve activity. In resting condition, the electrocardiogram (ECG) of healthy individuals exhibits periodic variation in R-R intervals. Heart rate (HR), a major determinant of Q, is controlled by factors intrinsic to the heart as well as by intrinsic neural and hormonal factors. The inherent rhythmicity of the heart, as established by its sinoatrial node, is regulated primarily by sympathetic and parasympathetic neurons emanating from cardioregulatory center in the medulla. The sympathetic cardioaccelerator nerves release norepinephrine at their endings, leading to increase of the HR during exercise. The parasympathetic vagus nerve releases acetylcholine, which tends to reduce HR [63-64].

Systolic Blood Pressure

The pressure in the arterial vessels is at the highest during level ventricular systole. Systolic bold pressure (SBP) is indicative of the force generated by the heart during ventricular contraction. Normal resting systolic pressure is about 120 mmHg [65].

Diastolic Blood Pressure

Diastolic blood pressure, the pressure in the arterial system during ventricular diastole, provided an indication of peripheral resistance. Normal resting diastolic blood pressure is approximately 80 mmHg. Dynamic, low-resistance exercise usually causes little or no change in diastolic blood pressure [65].

Human Body Temperature

As known that the traditional normal value for the human's oral temperature is 37±0.2ºC. Each part of body is at different of temperatures, and the magnitude of the temperatures differed between the parts always depends on the environmental temperature. The rectal temperature is representative the temperature at the core of the body and varies least with changes in environmental temperature. It also undergoes a regular circadian fluctuation of 0.5ºC. The temperature is lowest in the morning and during bed time but becomes highest in the evening and rises with activity such as exercise. At the time of women's ovulation, a rise of basal temperature causes an additional monthly cycle of temperature variation. With heavy exercise, the body temperature may rise 2ºC to 3ºC. The assessment of mean body temperature (MBT) must take into consideration both skin and core temperatures. This is typically accomplished by measuring the rectal temperature and a series of skin temperatures at various places on the body [66-67]. Mean body temperature is expressed by the following equation [67]:

MBT = (0.2 x skin temperature) + (0.8 x rectal temperature)

Respiration rate

Human respiration rate is measured when a person is at rest and involves counting the number of breaths for one minute by counting how many times the chest rises. Respiration rates may increase with fever, illness, or other medical conditions. When checking respiration, it is important to also note whether a person has any difficulty breathing. Inaccuracies in respiratory measurement have been reported in the literature. Average respiratory rate reported in a healthy adult at rest is usually given as 12-18 breaths per minute [64, 68].

Emotions in odors

Emotions and their expression are key elements in social interactions, being used as mechanisms for signaling, directing attention, motivating and controlling interactions, situation assessment, construction of self-and other's image, expectation formation, inter subjectivity, etc. It is not only tightly intervened neurologically with the mechanisms responsible for cognition, but that they also play a central role in decision making, problem solving, communicating, negotiating, and adapting to unpredictable environments. Emotion consists of more than its outward physical expression: it also consists of internal feelings and thoughts, as well as other internal process of which the person experiencing the emotion may not be aware, Individual emotional state may be influenced by kinds of situations, and different people have different subjective emotional experiences even response to the same stimulus [69-71].

Recently, a constellation of findings, from neuroscience, psychology, and cognitive science, suggests that emotion plays surprising critical roles in rational and intelligent behavior. When we are happy, our perception is biased at selecting happy events, likewise for negative emotions. Similarly, while making decisions, users are often influenced by their affective states. Reading a text while experiencing a negative valence emotional state that often leads to very different interpretation than reading the same text while in a positive state [72-73]. They are classified the different types of emotions elicited from the subjects through the physiological signals. They also described the different kind of emotions. The type of feature extraction technique, the method of eliciting emotions and the physiological signals used for classifying the emotions. Numerous experiments also showed that odors produce effects on cognition and behavior that are similar to those produced by emotional stimuli in other perceptual modalities. In addition, odor experiences have been shown to provoke changes in physiological parameters, such as heart rate or skin conductance, which are directly involved in the emotional responses [69, 72-74].

Numerous experiments also showed that odors produce effects on cognition and behavior that are similar to those produced by emotional stimuli in other perceptual modalities. In addition, odor experience has been shown to provoke changes in physiological parameters, such as heart rate or skin conductance, which are

directly involved in the emotional response [74, 75-77]. These effects are usually interpreted as an interdependence of olfaction and emotion on overlapping neural systems which has been recently confirmed with neuroimaging evidence. In this research, questionnaire procedure has been developed a conceptual model that proposes an modify of emotions The Geneva Emotion and Odor Scale (GEOS) described the subject affective feelings induced by five factor as follows [78]:

1. Pleasant feeling is mainly related to happiness and well-being, with a noteworthy association to ecstatic feeling as reflected by the term: feel good (รู้สึกดี) which has been used in this research.

2. Unpleasant feeling mainly related to disgust and irritation, but it also emphasizes other irritating feelings. In this research, selected words: feel bad (รู้สึกไม่ดี), uncomfortable (รู้สึกอึดอัด), disgusted (รู้สึกรังเกียจขยะแขยง) and frustrated (รู้สึกหงุดหงิด), stress (รู้สึกเครียด).

3. Sensuality reflects the role of olfaction in social interaction and, in particular, in sociosexual behaviors, that expressed by the terms ‘‘sensual,’’ ‘‘desire,’’ but selected word used in this research is romantic (รู้สึกเคลิบเคลิ้มรัญญวนใจ).

4. Relaxation is strongly associated with soothing effects, at the point that certain odors may induced meditative feelings. In this research, selected words including relax (รู้สึกผ่อนคลาย), serene (รู้สึกจิตใจสงบนิ่ง) and drowsy (รู้สึกง่วงซึม).

5. Refreshing is mainly associated with effects of stimulation and purification as well as physiological responses, which could be expressed by the terms: refresh (รู้สึกสดชื่น), and energetic (รู้สึกกระปรี้กระเปร่า).

Volatile oils properties

Volatile oil essential oil or ethereal oil is product of the secondary metabolism of plants. They are not actually oily, most of them are very light liquids that do not dissolve in water but evaporate instantly in the air. In general, volatile oils may be

present in many different parts of plants; wood, bark, leaves, stems, flowers, fruits, rhizomes etc. at concentration ranging from thousandths of a percent to one or several percent. Volatile oils are the life-blood of the plants, protecting it from bacterial and viral infections, cleansing breaks in its tissue and delivering oxygen and nutrients into the cells. In essence, they act as the immune system of the plant. That is why they are so essential to the plant, without them, plants could not survive. Volatile oils are much different from vegetable oils (also called fatty oils), such as corn oil, olive oil peanut oil, etc because pressing nuts or seeds produces fatty oils [1, 2, 79-82].

Chemical constituents of volatile oils.

Volatile oils are highly complex substances. Basic chemical structures of volatile oil molecules are made up primarily of carbon, hydrogen, and oxygen. The aromatic constituents of volatile oils are built from hydrocarbon chains (carbon and hydrogen atoms).which normally joined together in ring-like chemical structures. The chains are hold together by carbon atoms linked together. Oxygen, hydrogen, nitrogen, sulfur, and other carbon atoms attach at various points of the chain, to make up the different oils. The aromatic-ring structure of volatile oils is much more complex than the simple, linear carbon-hydrogen structure of fatty oils. Volatile oils also contain sulfur and nitrogen atoms that fatty oils do not have. Each volatile oil is made up of many different chemical components that combine in different ways to create specific oil. Some oils may have a few and others have hundreds. Different varieties of the same oil can have widely different therapeutic actions, depending on their chemistry. The chemical compounds in essential oils are broken down into two groups, hydrocarbons and oxygen based compounds. The oxygen-based compounds are Phenols, Alcohols, Esters, Aldehydes, Ketones, and Oxides. Hydrocarbons are mainly Terpenes (monoterpenes, diterpenes, and sesquiterpenes).

Phenols: This chemical group contains some of the most stimulating, bactericidal, and immune boosting essential oils. Phenols are water-soluble and evaporate more quickly than oils that do not contain phenols. Because of their strength, they can be irritating to the skin and possibly damaging to the liver. Compounds included in the phenol group are eugenol, thymol, carvacrol, gaiacol and australol.

Alcohols: These oils are generally uplifting, antiseptic, anti-viral, toning, non-toxic, and non-irritating. Alcohols are not water-soluble and evaporate quite slowly. Because they are less prone to oxidation, they will keep much longer. Common alcohol based chemicals are linalool, nerol, citronellol and patchoulol.

Esters: The esters sometimes have a trademark of fruity aroma. They are very balancing and calming. Esters are formed when acids and alcohols react with each other. Some common chemicals found in this group are linalyl acetate, neryl acetate and geranyl acetate.

Aldehydes: Oils containing this chemical group that are generally sedative and calming, with anti-inflammatory properties. These oils are usually quite strong and may cause skin irritation in some cases. The majority of the lemon scented oils are the aldehyde category. Some common chemicals found in this group are citral, geranial neral, and citronellal.

Ketones: The ketones consist of some of the most toxic elements of essential oils. Some of these include thujone and pulegone. The toxic chemicals in this group can cause epileptic seizures, convulsion, abortive effects, and mental confusion. However, not all ketones are toxic. The non-toxic ketones may aid in dissolving mucus, dissolving fats, and would healing. Some common chemicals in this group are jasmine, fenchone, carvone and menthone.

Oxides: In nature, Oils containing oxides are generally camphoraceous. They have wonderful expectorant properties. Some oils in this group contain chemicals, which may cause convulsant reactions, asarone and ascaridol. Some non-toxic oxides include cineol or eucalyptol, bisabolol oxide and linalol oxide.

Terpenes: Terpenes make up the hydrocarbon group. They can be broken down into three subcategories, Monoterpenes, Sesquiterpenes, and Diterpenes. Generally, terpenes are stimulating, anti-septic, and analgesic but may cause irritation to the skin. However, Sesquiterpenes have outstanding anti-inflammatory properties and is recently studied. Monoterpenes include limonene, pinene, camphene, sabinene and cadinine. Diterpenes are scarely found in volatile oils. The Sequiterpene group contains chamazulene, farnersol, valeranon and santalol.

Properties of selected volatile oils.

Essential oils used in the study have a range of chemical properties. The following section presents a summary of their properties.

Lavender Oil [83-84].

Botanical name: *Lavandula angustifolia* Mill.

Family: Labiatae/ Lamiaceae

Location:

Lavender grows widely along the Mediterranean coast but is also extensively cultivated for its fragrance in England and France

Extraction: Steam distillation of the fresh flowering tops

Color and Odor:

The essential oil is clear with a hint of yellow. It has a fresh sweet floral scent

Chemical constituents:

A typical chemical composition as reported [85]

- Linalyl acetate 44.16 %
- β–linalool 32.91 %
- Beta–caryophyllene 3.08 %

Toxicity to Animals:

Acute oral toxicity (LD50): ≥ 5000 mg/kg [Rat].

Acute dermal toxicity (LD50): ≥ 5000 mg/kg [Rabbit].

Chronic Effects on Humans: Not available.

Other Toxic Effects on Humans:

Slightly hazardous in case of skin contact (irritant, permeate), of ingestion

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Physiological effect:

The general properties of lavender oil are antibacterial, antifungal, carminative (smooth muscle relaxant), sedative, antidepressant, promoting wound healing and increasing the detoxification of enzymes associated with insecticide resistance. A number of researchers reported the sedative effects of lavender oil caused by the major components of linalyl acetate and β- linalool. These compounds can be rapidly absorbed through the body by inhalation with plasma level reaching a maximum peak in approximately 7 minutes after administration which can cause a depression of nervous system. Linalyl acetate has a narcotic action and linalool acts as a sedative. Diego and his colleagues found that individuals felt more relaxed and an improved mood after inhaling lavender oil. Moreover, an increase of mid frontal (F3, F4) alpha power on their EEG was found after inhalation of the oil. Motomura suggested that lavender decreased stress scores but increased Theta 1 (3.5-5.5Hz) brain wave activity and decreased Beta 1 (13.5-20 Hz) which is associated with relaxation [86-90].

Rosemary oil [83, 91]

Botanical name: *Rosmarinus officinalis* L.

Family: Labiatae

Location: Rosemary is cultivated in the Balkan stats England

Extraction: Steam distillation of the leaves

Color and Odor:

The essential oil is colorless and has a warm shap refreshing and camphorous aroma

Chemical constituents:

A typical chemical composition as reported [85]

- 1, 8-Cineole 21.36 %
- α- Pinene 21.18 %
- Camphor 13.86 %

Toxicity to Animals:

Acute oral toxicity (LD50): 5000 mg/kg [Rat].

Acute dermal toxicity (LD50): ≥ 5000 mg/kg [Rabbit].

Chronic Effects on Humans: Not available.

Other Toxic Effects on Humans:

Hazardous in case of skin contact (irritant), of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans: Not available.

Precaution:

It is highly stimulating action may not be suitable for people with epilepsy or high blood pressure. Avoid during pregnancy.

Physiological effects:

Rosemary oil is beneficial in the treatment of acne, baldness, rheumatic pain, circulatory block, and many more. In addition, rosemary oil has a pronounced action on the central nervous system and is wonderful for clearing the mind and mental awareness, with having excellent brain stimulant properties, as well as improving memory. The main chemical components of rosemary oil are α-pinene, camphor and 1, 8-cineole. The general properties of these substances include carminative, aromatic, antispasmodic, antidepressant, antimicrobial, astringent and stimulatory. In fact, scientists have learnt over years that the effects of rosemary oil can be rather extensive. In *vitro* study 1,8-cineole has been reported to have a stimulatory effect on the cerebral cortex of the rat. In animal studies, one hour after the addition of 0.5 ml rosemary oil per cage for evaporation, it was found that mice to be more locomotors actives. Furthermore, Graham investigates the influence of rosemary on the behavior of dogs. The diffusion of rosemary into the dogs' environment significantly encouraged more standing, moving than other types

of odor. The stimulatory effects could also be observed in human subjects after massage rosemary oil. A group of thirty fives volunteers their blood pressure and breathing rate increased and more attentive, alert and cheerful. In experiment using EEG (electroencephalography) found a significant decrease power of alpha wave over bilateral mid-frontal region. This finding therefore suggested that decreasing alpha power may relate to increasing level of alertness [91-97].

Citronella [83]

Botanical name: *Cymbopogon nardus* Rendle

Family: Poaceae

Location: Native in Srilanka , South Asia , and South East Asia

Extraction: Steam distillation

Color and Odor: The essential oil is clear white

Chemical constituents:

A typical chemical composition as reported [85]

- β-Citronellal 39.07 %
- Cis-Geraniol 24.81 %
- β-Citronellol 10.77%
- D- Sylvestrene 6.64 %

Toxicity to Animals:

Acute oral toxicity (LD50): 7200 mg/kg [Rat].

Acute dermal toxicity (LD50): 6700 mg/kg [Rabbit].

Chronic Effects on Humans:

The substance is toxic to lungs, mucous membranes.

Other Toxic Effects on Humans:

Very hazardous in case of skin contact (irritant), of ingestion. Hazardous in case of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans: Not available.

Precaution:

Non-toxic non irritant a few cases of sensitization have been reported

Physiological effect;

The sedative effect of citronella was confirmed in experimental animals by Jaeger *et al*. Their research found that under standardized experimental conditions, the motility of female mice was reduced from arbitrarily graded from 100% for untreated animals to 50.18% by citronellal. In the serum of animals exposed for one hour showed the concentration of the citronellal of 2.53 ng/mL**.** In addition, citronella spray collar significantly reduce baking in 30 dogs. Furthermore, in human study of Saeki and Shiohara, they demonstrated that after inhaling citronella treatment R-R interval on the electrocardiogram was increased whereas blood pressure was decreased combining simultaneously with calm and relax emotions [98-100].

Jasmine oil [83]

Botanical name: *Jasminum sambac* (L.) Aiton.

Family: Oleaceae

Location: Jasmine is originated in China and India it is cultivated in Egypt France Italy Morocco and Turkey

Extraction: Solvent extraction of the flowers

Color and Odor: The absolute is a deep reddish brown color and has a sweet exotic slightly heady aroma with a hint of musky tones

Chemical constituents:

A typical chemical composition as reported [85]

- Benzyl acetate 26.09 %
- β – linalool 11.22 %
- Benzyl propionate 9.65 %
- Methyl anthranilate 8.54 %

Toxicity to Animals:

LD50: Not available.

LC50: Not available.

Chronic Effects on Humans: Not available.

Other Toxic Effects on Humans:

Hazardous in case of ingestion. Slightly hazardous in case of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans: Not available.

Physiological effect:

Jasmine is supposed to be a stimulating on human attention. The basic level being that of alertness which ranges from sleep to wakefulness. In 1991, Tsuchiya *et al*. reported the effects of jasmine aroma on mice sedated using pentobarbital. In human study, Hongratanaworakit shown that after applied jasmine oil topically to the skin at abdomen of 40 volunteers. Compared with placebo, jasmine oil caused significant increases of breathing rate, blood oxygen saturation, and systolic and diastolic blood pressure, which indicated an increase of autonomic arousal. At the emotional level, subjects in the jasmine oil group rated themselves as more alert, more vigorous and less relaxed than subjects in the control group. Furthermore, Sugano and Nakagawa *et al*. found that jasmine odor has an effect on increasing beta wave activity which is a stimulating effect on the brain, Methyl jasmonate, major

component of jasmine oil, inhibited the enhancement of alpha and theta waves which seemed to show a stimulating effect of the odors [15, 101-103].

Related reviews

Effect of aromas on brain wave

In general, the effects of aromas evaluated by EEG measurements showed brain wave responses in amplitude and frequency. Aromas produced the cortical brain wave activity responses involving alpha, beta, delta, and theta waves. It has been shown that beta wave is dominant when people engaged in reading, concentrated deliberation, highly emotional and other tense mental states. In contrast, alpha wave is inhibited in the same situation. Likewise, alpha wave is dominant in mentally relaxed state. It could be suggested that, brain waves consistently reflect human levels of consciousness, psychological states and degree of arousal. The brain waves of subjects exposed to the four types of aromas, i.e., eucalyptus, lavender, spiced apple and odorless solvents as a control have been recorded by Lorig and Schwartz. The results demonstrated that all aromas were linked with different alpha and theta wave distributions. A study conducted at University of Occupational and Environmental Health, Kitakyushu, Japan used variations in EEG readings as indices for the measurement of aroma effects. The aromas of lavender, cineol, jasmine, and sandalwood were explored. A relaxing effect (increase of alpha wave activity) was found upon presentation of lavender, cineol, sandalwood. In contrast, a stimulating effect that increases beta wave activity was found upon presentation of jasmine odor. Nakagawa *et al.* reported that methyl jusmonate and cineol aromas inhibit the enhancement of alpha and theta waves, resulted in a stimulating effect of the odors. On the other hand, jasmine lactone odor enhanced the quantity of alpha and theta waves which probably indicated a relaxing effect of this odor.

Several studies have shown influences of odor pleasantness and familiarity on changes of the spontaneous EEG. For instance; Tonoike *et al.* reported that pleasant odors increased the alpha activity, while unpleasant ones decreased it. In Lorig's study, effects of lavender and jasmine odor on electrical brain activity were revealed that having changes in the alpha, beta, and theta bands in subjects rated as pleasant, while showing distinct patterns in subjects rated as unpleasant. As pleasant odors

induced deeper inhalations and exhalations more than unpleasant odors, so this altered breathing increased alpha activity in the band. Furthermore, Ekman *et al*. found negative emotions were related to right frontal lobe while happiness excited more the left frontal lobe activity, Bensafi found the output of alpha waves was significantly reduced in the right compared with the left frontal brain region when volunteers were stimulated with a pleasant odor (vanilla). In conclusion, the effects of aromas on electroencephalogram are depending on two factors of characteristic of essential oil and pleasantness [102, 104-110].

Effect of aromas on heart rate

Heart rate is the most common psychophysiological measurement of heart activity. A faster heart rate is often caused by stress. The heart may race and pound when people feel anxious, for example, depression, another kind of stress, perhaps leads to a decrease in heart rate. The heart is innervated by the ANS. The ANS is subdivided into the sympathetic nervous system (SNS) and the parasympathetic nervous system (PSNS). Those are completely different in functions. The PNS reduces the activities of the heart and particularly influences heart rate, whereas the SNS increases the activities of the heart and particularly affects the pumping function [53-54, 63]. A study by Yamaguchi also used the changes of heart rate to measure effects of lemon and rose aromas. Lemon aroma caused an increase in heart rate, whereas rose aroma led to a decrease. This finding probably indicates that lemon aroma has a stimulating effect (an increase of heart rate), by contrast, rose aroma possesses a sedative effect (a decrease of heart rate). In a similar investigation of Kikuchi and co-workers, lemon aroma enhanced the deceleration of the heart rate, indicating a stimulating effect. Alternatively, rose aroma suppressed it which is likely represented a sedative effect. Nagai *et al*. showed that sweet fennel oil suppressed the deceleration of heart rate as well. Hongratanaworakit *et al*. investigated the effects of sweet orange aroma on human behavior and detected changes of heart rate in response to olfactory stimulation. They reported that sweet orange aroma caused significant increases in heart rate and subjective alertness after inhalation. These findings seemed to show a stimulating effect of sweet orange oil. Shiina *et al.* assessed the effect of lavender aromatherapy on coronary circulation by measuring coronary flow velocity reserve (CFVR) and serum cortisal (that makes stress hormone). After aromatherapy,

CFVR showed a significant increase on induction of vasodilatation and a decrease on serum cortisal. This research suggested that lavender had a relaxating effect and might have an effect on coronary circulation. In the research by Brauchli *et al*. they reported that a pleasant and an unpleasant odor presentations affected an autonomic variable, that is, heart rate. An increase of heart rate was observed during the valeric acid presentation. In contrast, a decrease of heart rate was found during the phenylethyl alcohol presentation. Phenylethyl alcohol was rated as pleasant, while valeric acid was judged as unpleasant. Thus, the pattern of changes in the heart rate revealed differences between stimulant aromas and sedative aromas connecting to two factors of characteristic of essential oil and pleasantness [24, 111-114].

Effect of aromas on blood pressure

Blood pressure is one of the most frequently measurement for physiological variables. It is used as a general index of cardiovascular function and health. The maximal, or systolic blood pressure occurs when the ventricle of the heart contracts. Following the period of cardiac contraction, there is relaxation of the ventricle, during the time that blood pressure is hold at a minimum; a measurement at this time yields diastolic blood pressure. Blood pressure is regulated by various factors such as blood volume and peripheral resistance. In general, diastolic blood pressure varies mostly with peripheral resistance, whereas systolic blood pressure is related to both peripheral resistance and stroke volume. Blood volume is much less familiar than blood pressure. It refers to the amount of blood that is present in a certain portion of body tissue at a given time. If blood volume is low, blood pressure is reduced. More blood is pumped by the heart, more blood vessels are constricted by the higher level of blood pressure. All blood vessels inside body, except the capillaries, are innervated by nerve fibers from the SNS alone. This is controlled via the vasomotor center, which is located in the reticular substance of the brain (lower pons and upper medulla). The hypothalamus of the brain can exert powerfully inhibitory or excitatory effects on the vasomotor center. Thus, blood pressure was recorded as an indicator of the arousal level. A study conducted at the Royal Sussex County Hospital showed that foot massage with essential oil of lavender lowered blood pressure as well as heart and respiratory rates of patients in an intensive care unit. Transdermal absorption of sandalwood oil and one of its main components, α-santalol led to a trend towards a

larger decrease of systolic blood pressure as compared to that of the placebo group. Furthermore, effects of chiral fragrances on human blood pressure and self-evaluation were explored by Heuberger *et al*. In their studies, chiral fragrances (enantiomers of limonene and carvone) caused increases in blood pressure, subjective restlessness and alertness. These findings are likely to represent a stimulating effect of these fragrances. Hongratanaworakit *et al*. have demonstrated that a ylang-ylang oil exhibited a harmonizing effect. Inhalation of the ylang-ylang oil led to a decrease of blood pressure and an increase in subjective attention. In addition, transdermal absorption of the mixed oil of bergamot and lavender oils caused a significant decrease of blood pressure .This finding points resulted in a decrease of autonomic arousal [13, 23,115-118].

Effect of aromas on skin temperature and breathing rate

Individual cognitive and emotional psychological states may cause changes in skin temperature and respiratory rate. For example, events that activate the sympathetic nervous system, leading to a stress response, may reduce skin temperature but increase respiration by reducing peripheral circulation. Also, survived animal often cope the stress which lead to a collection of physiological response: lower skin temperature, increasing heart rate and brerating, increasing muscle tension; these are accompanied with psychological change such anxiety etc. The opposite events occur when the stressful situation passed. Parasympathetic activation returns blood flow to the peripheral, resulted in the increase of peripheral skin temperature, decrease of breathing rate and induce relaxation. A study conducted by Hongratanaworakit *et al*. have demonstrated that after transdermal absorption of ylang-ylang oil, caused a significant increase of skin temperature. In addition, transdermal absorption of the mixture of bergamot oil and lavender oil caused the significant decrease of breathing rate and the increase of skin temperature. Furthermore, Hongratanaworakit *et al*. have presented that transdermal absorption of sweet orange oil, led to a significant decrease of breathing rate. But significant effects of the sweet orange oil on skin temperature had not been reported [13, 25, 83].

Effect of aromas on emotions.

In most cultures, odor has is a powerful elicitor of emotions. In the last few decades, a growing scientific literature has documented various emotional effects of odors. By using various approaches, research investigating the relation between odor and affective phenomena showed, for example, that odor experience is inextricably linked to odor hedonic tone. Thus, odor is likely to influence mood such that pleasant odors tend to induce positive moods, whereas unpleasant odors tend to induce negative moods. Numerous experiments also showed that odors produce effects on cognition and behavior that are similar to those produced by emotional stimuli in other perceptual modalities. In addition, odor experiences have been shown to provoke changes in physiological parameters, heart rate and skin conductance, which are directly involved in the emotional responses [119-121].

Sattely-Miller assessed whether the daily use of pleasant smelling colognes could elevate mood in men by using the Profile of Mood States (POMS) questionnaire which can be divided into six factors: tension-anxiety, depression-dejection, anger-hostility, vigor-activity, fatigue-inertia. The first two days of the study provided the baseline information for each participant. For both the baseline and the remainder of the study, the POMS was completed twice each day. Statistical significance was found for the vigor factor, with the fragrance condition having significantly higher scores than the placebo condition. Lehrner *et al*. examined the ability of orange odor to reduce anxiety and improve mood in dental patients while waiting for a dental treatment. Participants were assigned to either a control condition (where they waited with no odor present) or to a scent condition (where ambient orange scent was diffused into the waiting room). Compared to the control group, the orange scent group reported a lower level of anxiety, a more positive mood, and a higher level of calmness. Burnett and her colleges presented about participants with the scent of lavender, rosemary, or water. Both rosemary and lavender scents associated with lower mean ratings on the fatigue-inertia subscale of the Profile of Mood States, that related to the control group [122-124].

CHAPTER III

MATERIALS AND METHODOLOGY

Location and setting

The study was conducted at Salaya Stem Cell R&D Project, 6th Floor Panyawattana Building, National Institute for Child and Family Development, Mahidol University, Salaya Campus. A temperature of laboratory was set at 25 ^{0}C with a relative humidity between 50-65%. All measurements were taken in a quiet room. The experiments were performed between 8.00-12.00 a.m. to minimize circadian variation [125].

Research Design

This study was an experimental research. An A-B design has been used, so that each individual session consisted of two trails. This design was chosen because, with olfactory stimulation, the times court of stimulatory effects is unknown, which might make results obtained from other designs, such as A-B-A, difficult to interpret [126].

Subjects

Eighty subjects were recruited from Mahidol University, Thailand.

Inclusion criteria

1. Thai native speakers, aged ranging from 18 to 35 years.
2. The handedness of the participants was assessed with the Edinburgh Handedness Inventory. The degree of the right handedness of the participants were assessed in ten items, including writing, drawing, throwing, scissor-cutting, tooth brushing, knife-cutting (without fork), spoon, broom, striking a match, and opening box lid. The participant was instructed to make a "+" on which hand he would prefer to use for each action. The participant then was instructed to mark a "++" when the preference was so strong that he never used the other hand unassisted. If, In case, the participant did not have any preference, he was further

instructed to mark a "+" for both hands. The numbers of "+ "marked for each hand were totaled. Then, a handedness index was calculated to be the difference of the numbers of "+ " 's between the right and left hands divided by the total number of "+ "'s for both hands. A handedness index of 1.0 indicated completely right handed, -1.0 corresponded to completely left handed, and 0 suggested ambidextrous. The participant was also be asked which foot was preferred for kicking, which eye was preferred when only using one eye, and whether both parents were right handed. The handedness index was then be calculated.

$$\text{Handedness index} = \frac{\Sigma\,(R) - \Sigma\,(L)}{\Sigma\,(R) + \Sigma\,(L)}$$

Whereas Σ (R) is the summation all item of right hand and Σ (L) is the summation all item of left hand.

Participants who strongly in left handed were excluded from experiment [127].

3. Before the experiment, subjects have been tested the normal sense of smell by "n-butyl alcohol method test". This protocol measured the lowest concentration of a stimulus that can be distinguished between n-butyl alcohol and water. Normal subjects could separate two odors at concentration lower than Step 6 (5.48×10^{-3} v/v) of n-butyl alcohol in water [51].
4. Subjects should not have otorhinolaryngologic, upper respiratory infection, neurological diseases, hypertension and cardiovascular disease.
5. Normal blood pressure (systolic should not be higher than 140 mmHg, diastolic should not be higher than 90 mmHg)
6. Heart rate should not be higher than 90 with normal rhythm.
7. Body mass index was between 18 and 25 kg/m^2
8. Subjects did not have history related to neurological illness, epilepsy and loss of consciousness longer than 30 minutes.

9. Did not taking CNS medication and presently taking recreational drugs
10. Non –smoker.

Exclusion criteria

1. Subjects had colds and physical conditions that may affect to the sense of smell.
2. Women presently have menstruating.
3. Drowsiness before experiment.
4. Taking caffeine and alcohol two hours before the test.
5. Abnormal brain wave detected by electroencephalogram.

Sample size determination

Determination in this study was calculated from the previous study of lavender oil that affected to autonomic nervous system, blood pressure, heart rate and breathing rate [12]. The sample was calculated from a dependent group formula [128].

$$N = \frac{2 \times (Z_\alpha + Z_\beta)^2 \times sd^2}{d^2}$$

α = 0.05 (Two sided) Z_α = 1.96

β = 0.20 (Two sided) Z_β = 1.28

sd = The difference of standard deviation diastolic before and after lavender inhalation

d = The difference of mean diastolic before and after lavender inhalation

N = Group of subjects

$$N = \frac{2 \times (1.96 + 1.28)^2 \times (1.68)^2}{(1.81)^2} = 18$$

To account for the expecting drop outs during experiment and to ensure the study confidence, more than 10% of subjects are recruited. There are 20 subjects recruited by announcement.

Sampling technique

Purposive sampling technique has been chosen for subject recruitment. The odor familiar questionnaire was required for selecting an essential oil for individual subject. Subjects should not have experience in distress after inhalation of the essential oil. Before the experiment, they were asked to inhale sweet almond oil and essential oil to estimate the pleasantness of the smell on a five-point Likert scale. The participants, who indicated oil pleasantness within the target level range of 2-4 were chosen to participate in this study.

Essential oil

Sample collection

Four types of natural essential oils have been used in this study.

1. Lavender oil (*Lavandula angustifolia* Mill.)
2. Rosemary oil (*Rosmarinus officinalis* L.)
3. Jasmine oil (*Jasminum sambac* (L.) Aiton)
4. Citronella oil (*Cymbopogon nardus* Rendle)

All essential oils obtained from Thai China Flavors and Fragrances industry. These essential oils are widely used in spa and cosmetic products.

Essential oil Analysis

The oil composition was identified by gas chromatography /mass spectrometry (GC/MS) (Thermo Finnigan model Trace GC Ultra equipped with Finnigan DSQ MS detector, USA). The constituents of the oil were identified matching their mass spectra and retention times indicated with NIST05 MS library, and the percentage compositions were computed from GC peak area [129-130].

Essential oil delivery

In terms of odor delivery, the mixture of essential oil and base oil (sweet almond oil) at 10 % v/v concentration was delivered from an oxygen pump system through a plastic tube via a face mask that permitted selective routine air flow (2L/min).

Instruments and other Supplies

Screening session

1. Health status (Appendix D).
2. Edinburge Handedness Inventory test (Appendix E)
3. Score sheet for odor test **(**Butanol Threshold**)** (Appendix F)
4. Odor familiarity test (Appendix G)

Autonomic nervous system and emotions measurement

1. Life scope 8 Bedside monitors (Nihon Kohden, Japan)
2. Comfortable armchair
3. Case record autonomic nervous form (Appendix H)
4. 70% alcohol
5. ECG conductive adhesive electrodes **(**Medi-Trace 230)
6. Emotions recording questionnaire (Appendix I)

Electroencephalographic Recording

1. Nu Amps amplifier and head box
2. 15 inch USB cable (connection EEG acquisition computer–Amplifier)
3. Serial cable (connection Stimulation computer–Amplifier)
4. SCAN 4.3 software
5. CPU and monitor for SCAN software (EEG acquisition)

6. Comfortable armchair
7. EEG Quick Cap with 40 channels (including HEOG/VEOG, reference and ground electrodes), Ag/AgCl electrodes.
8. Disposable blunt needles and syringes
9. Electro-gel (Siriraj Hospital Pharmacy, Bangkok)
10. Ivory liquid (OMNIPEP)

(A) (B)

Figure 5 ANS instruments (A) Life s cope 8 Bedside m onitor (B) Comfortable armchair

(A) (B) (C)

Figure 6 EEG instruments (A) EEG acquisition (B) NuAmps Amplifier and Cables (C) EEG Quick caps

Protocol

Screening protocol

1. The research project was announced to public via the leaflets and fitters.
2. The criteria for subjects who interested in this research are these followings:
   2.1. Before the screening test, participants needed to sign consent form and attended the tutorial consisting of the instructions which was informed in Thai (Appendix B).
   2.2. The personal health status has been assessed by questionnaire. Measurements of weight, height and blood pressure were also required(Appendix D).
   2.3. In this study, Edinburgh Handedness Inventory test has been chosen to evaluate the handedness of the participants(Appendix E)
3. The olfactory ability evaluation has been performed by using these following steps: (Appendix F)
   3.1. Butanol and water solutions at various concentration ranged from 0 to 11 were prepared and kept in the bottles.
   3.2. Subjects were asked to identify the bottle containing the odorant, at initial concentration level 9.

3.3. After each correct response, the concentration of butanol was decreased by a factor of 3 (level 10, 11).

3.4. After each incorrect response, the concentration of butanol was then increased by a factor of 3 until the participant either achieved 5 correct responses or failed to identify the bottle with 4% butanol.

3.5. The detection threshold was recorded as the concentration at which the participant correctly identified the butanol on 5 consecutive trials. The score then linked the patient's threshold to a normal subject population.

4. To evaluated odor preference in this step, individual participant had to sniff a paper dropped with following odors(Appendix G):

4.1. Lavender oil

4.2. Rosemary oil

4.3. Jasmine oil

4.4. Citronella oil

The next appointment was confirmed at the end of each experiment. One day before the experiment date, researcher needed to contact participants by phone to confirm the experiment date. Before the experiment starting, subjects needed to shampoo their hair. However, hair spray, antiperspirants and perfumes are not allowed. Additionally, participants should avoid alcohol and caffeinated drinks as well as smoking. They should not be fatigued or drowsy during the experiment date.

Autonomic nervous system measurement

Autonomic parameter recording

Four ANS parameters: skin temperature (ST), breathing rate (BR), heart rate (HR) and blood pressure (BP), were recorded simultaneously in real time. All parameters were measured using Life scope 8 Bedside monitors with the participant seated in a semi-reclining chair in the room with quiet, air-conditioned (24° ± 1°) and 40-50% humidity. All tests were performed between 08.30 AM and 12.30 AM to

minimize circadian variation of the autonomic nervous system. To avoid mutual distraction during the test, each participant was tested separately. The room was ventilated with fresh air for at least 15 minutes before the next participants. Following parameters were recorded (Appendix H).;

1. Heart rate and breathing rate measurement (every 1 minute). Connected the electrode lead in three positions (Modified Lead I, II, III) included left infraclavicular fossa, right infraclavicular fossa and left anterior axillary line below the bottom rib. In this position, respiratory measurement is influenced by movement of chest and abdomen on the left infraclavicular fossa and the left anterior axillary line below the bottom rib as shown in figure 7a
2. Blood pressure measurement (every 2.5 minutes). Systolic Blood Pressure (SBP) and Diastolic Blood Pressure (DBP) were measured on the left arm as shown in figure 7a.
3. Skin temperature measurement (every 1 minute). The sensor was placed in middle of the back of a non-dominant hand and fixed with non-caustic adhesive tape as shown in figure 7a.

Emotions recording

In this research, modify questionnaire procedure developed a conceptual model that proposes an aspect of emotions. The Geneva and odor scale (GEOS) The described the subjective affective feelings induced by 5 factors as follows:

1. Pleasant feeling related mainly to happiness and well-being, with a noteworthy association to ecstatic feeling as reflected by the term: "feel good (รู้สึกดี)" that has been used in this research.
2. Unpleasant feeling not only related to disgust and irritation but it also emphasizes other irritating feelings. In this research, mainly words including "feel bad (รู้สึกไม่ดี)", "uncomfortable (รู้สึกอึดอัด)", "disgusted (รู้สึกรังเกียจขยะแขยง)", "frustrated (รู้สึกหงุดหงิด)" and "stress (รู้สึกเครียด)" have been selected.

3. Sensuality reflects the role of olfaction in social interaction and, in particular, in sociosexual behaviors, that expressed by the terms ‘‘sensual’’ and ‘ ‘desire”. The appropriate word used in this study was “romantic (รู้สึกเคลิบเคลิ้มรัญญวนใจ)”.

4. Relaxation strongly associated with soothing effects at the point that certain odors may induce meditative feelings. This research used words including “ relax (รู้สึกผ่อนคลาย)”, “serene (รู้สึกจิตใจสงบนิ่ง)” and “drowsy (รู้สึกง่วงซึม)”.

5. Refreshing mainly linked with stimulation and purification effects as well as physiological responses that could be expressed by the terms “refresh (รู้สึกสดชื่น)” and “energetic (รู้สึกกระปรี้กระเปร่า)”.

The questionnaire has been verified by advisor, co- advisor, specialist in Thai interpreter and physiologist. The 100 millimeter visual analog scale has been chosen in this study to assess emotion condition (Appendix I). The measure reliability were done by 20 participants in preliminary study and calculated for Cronbach’s α value. The measure with Cronbach’s α value = 0.752

(A) (B)

Figure7 Autonomic parameter recording

(A) The electrodes connected to subject (B) Data collection during experiment

Electroencephalographic Recording

The set of 31 electrodes with 1 additional ground which was placed according to the international 10-20 system at FP1, FP2,FZ, F3, F4, F7, F8, FT7, FC3, FCZ, FC4, FT8, T3, T4, T5, T6, TP7, TP8, C3, CP3, C4, CZ, CPZ, CP4, P3, P4, PZ, Ol, O2 and O Z. Both mastoids were used as the recording reference (average of both mastoids, Al + A2/2). The electro-oculogram (EOG) was monitored with 4 electrodes placed in both external acanthi (HEOL and H EOR), left supraorbital (VEOU) and infraorbital (VEOL) regions. Electro-Caps were made of an elastic spandex-type fabric with recessed, silver/silverchloride (Ag/AgCl) electrodes attached to the fabric. Electrode impedances were set below 5 k Ohms. The recording system used in this study was Acquire Neuroscan version 4.3 from Neurosoft, INC. The online filter was set to a bandpass with the low pass is equal to 70 Hz and the high pass is equal to DC. A/D rate was 500 Hz and Gain was set at 19. Notch filter was open at 50 Hz. The relative power spectrum of the respective frequency bands derived from Fast Fourier Transformation (FFT) were expressed as follows: The relative power spectrum of the respective frequency bands derived by Fast Fourier Transformation (FFT) were expressed as follows: Delta (0 –3.99 Hz), Theta (4–7.99 Hz), Alpha (8–12.99 Hz), Alpha1 (8–10.99 Hz), Alpha2 (11–12.99 Hz) and Beta (13–29.99 Hz) shown in figure 8.

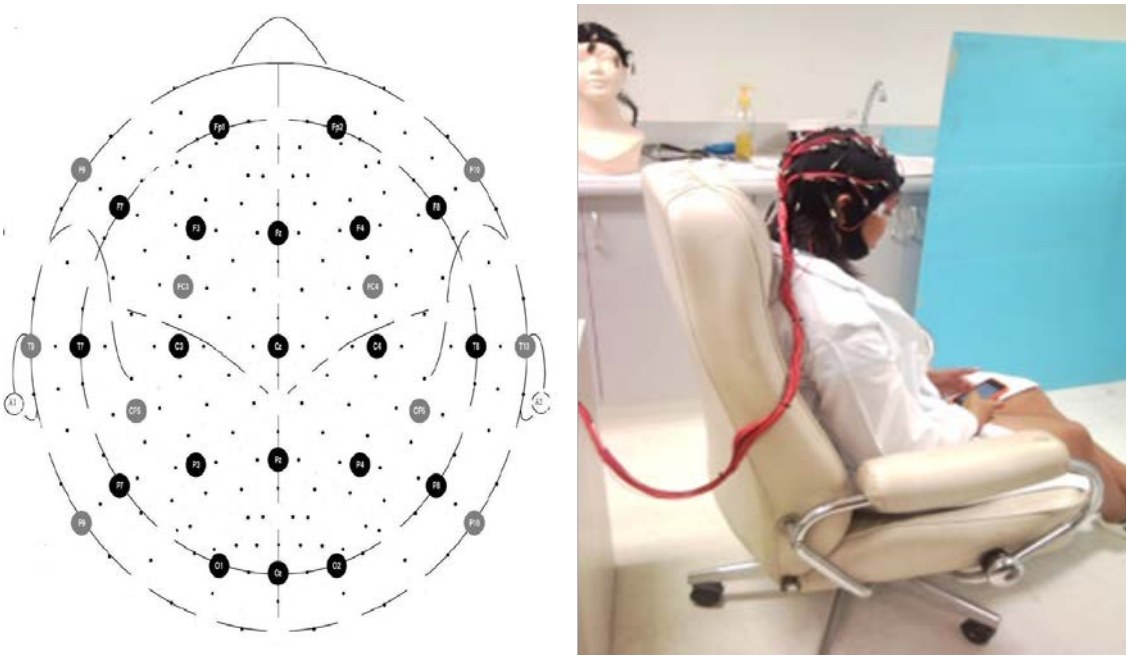


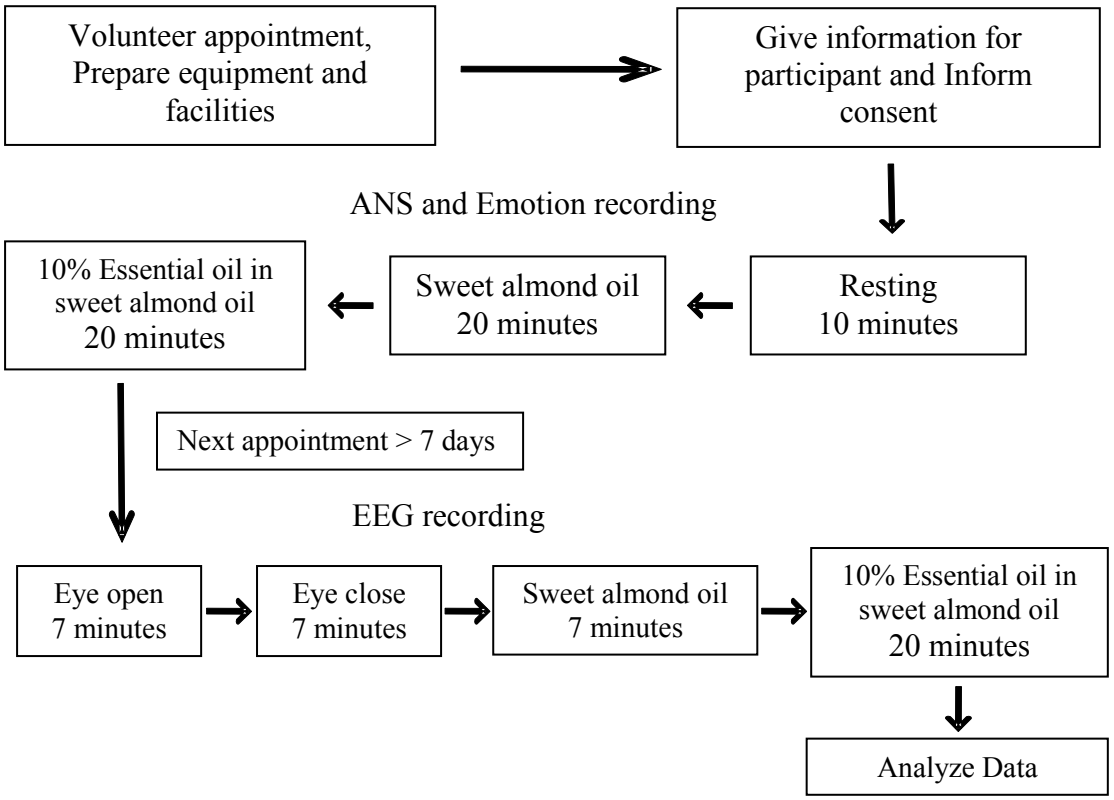
Figure 8 Electrode placements according to the 10-20 system

Experiment procedure

1. Before the experiment, subjects did not allow to use hair spray, antiperspirants and perfumes. In addition, alcohol, cigarettes and caffeinated products should be avoid. Participants should not be fatigued or drowsy during experiment date.
2. Before the ANS recordings, participants needed to sign consent form and attended the tutorial consisting the instructions which informed in Thai (Appendix B).
3. Participant was sitting in a comfortable chair separately from the ANS acquisition unit with the room temperature at 24-26 C, relative humility at 50-65% and silent.
4. As soon as subject feels comfortable, the first measurement of emotions and ANS activity was recorded 10 minutes.
5. To measure the second ANS activity and emotions, undiluted sweet almond oil was applied for 20 minutes.

6. To measure the ANS activity and emotions at the third trial, 10 % v/v sweet almond oil was applied for 20 minutes.
7. ANS and Emotions recordings have been analyzed.
8. Participants were required to measure their brainwave again after the experiment no less than 7 days.
9. The EEG experimental conditions were the same as autonomic nervous system experiment.
10. Baseline EEG recording was done with both eyes opened and eyes closed for 7 minutes respectively. After that. Finally 10 % v/v essential oil in sweet almond oil was inhaled.
11. At the third trial, participants would be inhaled undiluted sweet almond oil for 7 minutes.
12. Finally, participants would be inhaled 10 % v/v essential oil in sweet almond oil for 7 minutes. The procedures are sequenced as in a following map.

The procedures are sequenced as in a following map.



Data analysis [131]

Experimenter had collected results from all experiments and analyzed data using statistics;

1. Descriptive Statistics explain general data such as frequency, percentage, mean and standard deviation of participant's data, ANS data and EEG power.
2. Komogorov Smirnov Goodness of fit test performs to distribution of the data.
3. Effects of essential oil on physiological and emotions before and after were analyzed by paired *t*-test (parametric) and Wilcox sign rank test (non parametric).
4. Spearman rank correlation was performed to determine the association between physiological change and emotions.

Ethical Review

The present study was approved by the Ethical Review Committee for Research Involving Human Research Subjects, Health Science Group, Chulalongkorn University, Permissions no. COA NO. 009/2011(Appendix A). Purposes and procedures of the study were clearly explained to the participants. Informed consent from participants was obtained. The respondents have been informed that they are free to withdraw any time throughout the experiment. All data will be kept confidential.

Limitations

1. Emotions stimulated by odors can be varied according to individual experience with the odors.
2. The pattern of brain wave could be changed if participants show the uncomfortable signs such as eye blink, and emotional changing during the experiment

Expected Benefit & Application

1. This study will be useful for selecting the most appropriate essential oil for certain outcomes.For example, the best essential oil for fleshing condition can increase Alpha wave and heart rate. Moreover, the best essential oil for relaxation must decrease heart rate but increase Beta waves. Finally, cognitive conditions can be induced by the essential oil that increases Theta wave.
2. The information is expected to provide information related to safety and awareness of essential oils that increase blood pressure for people who have history about high blood pressure. Furthermore, drives should avoid essential oils for relax and sleepy in this car.
3. This protocol could be applied for further researches in order to study effects of other essential oils and their mixture on physiological effect and emotions.

CHAPTER IV

RESULTS

Lavender oil

Lavender oil components

The oil composition was analyzed by gas chromatography /mass spectrometry (GC/MS), Thermo Finnigan model Trace GC Ultra equipped with Finnigan DSQ MS detector, USA). The oil constituent was identified with mass spectra whereas retention times from NIST05 MS library and the percentage compositions were computed from GC peak area. Two main components of lavender oil were comprised of linalyl acetate (32.46%) and linalool (31.91%) (Appendix J).

General characteristics of participants

Twenty participants (10 males and 10 females) aged between 18 and 35 years (mean age 23.25 $\pm$ 4.52 years) with normal body mass index (mean 20.86 $\pm$ 1.91) were enrolled in this study. As a number of studies showed differences in activity between the left-handed and right-handed subjects during olfactory tasks; thus, only right-hander were tested. Handedness was tested using Edinburgh Handedness Inventory scale. The subjects were then screened for a normal sense of smell by the n-butyl alcohol method test (mean score 10.00 ± 0.77). A summary of the demographic data of the participants is presented in Table 2.

Table 2 Demographic data for the lavender inhaling participants.

Parameters	Number	Minimum	Maximum	Mean	SD
Age	20	18	38	23.25	4.52
Height(cm)	20	152	177	167.43	6.82
Weight (kg)	20	46	71	58.57	6.38
Body Mass Index (kg/m^2)	20	17.85	24.71	20.86	1.91
Smell test	20	9	11	10	0.77

Autonomic Nervous System Parameters

The mean and Standard Deviation (SD) values of autonomic parameters in the experiment are presented in Table 3 and Figure 9. The data were compared on various autonomic parameters during resting and inhaling sweet almond oil. Subjects had significantly decreased heart rate and breathing rate (p-value <0.05) during the sweet almond oil treatment when compared to those during resting. Moreover, when subjects inhaled the lavender, the systolic and diastolic blood pressures, heart rate and skin temperature were significantly decreased compared to those of sweet almond oil inhalation. However, a significant decrease was found on respiratory rate (RR), when compared between resting state and base oil state, whereas its increase was shown significantly in volunteer during lavender inhalation.

Table 3 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and lavender oil inhalations.

Minute	**Systolic Blood Pressure (mmHg)**						***p*-value R and SO**	***p*-value SO and LO**
	Rest		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	109.98	8.95	111.05	10.21	109.36	8.45		
10	109.84	10.93	109.85	9.76	108.44	8.41		
15			110.30	8.93	107.17	7.83		
20			109.90	9.78	107.06	8.36		
Average	109.91	9.74	110.27	9.51	108.00	8.41	0.588	0.000*
Minute	**Diastolic Blood Pressure (mmHg)**						***p*-value R and SO**	***p*-value SO and LO**
	Rest		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	70.12	7.81	71.15	8.85	69.15	8.57		
10	68.52	9.71	69.60	9.39	68.40	9.30		
15			69.70	8.44	68.40	7.39		
20			70.60	9.71	68.15	8.43		
Average	69.32	8.76	70.26	8.96	68.52	8.43	0.527	0.000*
Minute	**Heart rate(beat/min)**						***p*-value R and SO**	***p*-value SO and LO**
	Rest		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	72.18	12.22	69.51	12.99	67.11	10.26		
10	70.22	11.16	68.61	9.39	66.16	10.48		
15			67.58	13.32	65.14	11.62		
20			68.00	12.09	64.33	11.12		
Average	71.20	11.69	68.43	12.86	65.68	10.73	0.001*	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), lavender oil (LO)

Table 3 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and lavender oil inhalations (Continue).

Minute	Respiratory rate (bpm)						*p*-value R and SO	*p*-value SO and LO
	Rest		*Sweet almond oil*		*Lavender*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	18.45	9.22	15.50	2.63	15.91	5.59		
10	18.43	9.46	15.58	3.11	16.33	7.86		
15			15.86	3.16	16.42	6.81		
20			15.87	3.16	16.55	6.81		
Average	18.44	9.34	15.70	2.91	16.36	6.71	0.029*	0.148
Minute	**Skin temperature (°C)**						***p*-value R and SO**	***p*-value SO and LO**
	Rest		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	31.11	1.55	31.22	1.81	31.1	1.90		
10	31.17	1.73	31.37	1.67	31.09	1.84		
15			31.17	2.35	30.99	1.97		
20			31.26	2.11	30.81	2.16		
Average	31.14	1.64	31.25	1.96	31.00	1.94	0.296	0.001*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), lavender oil (LO)

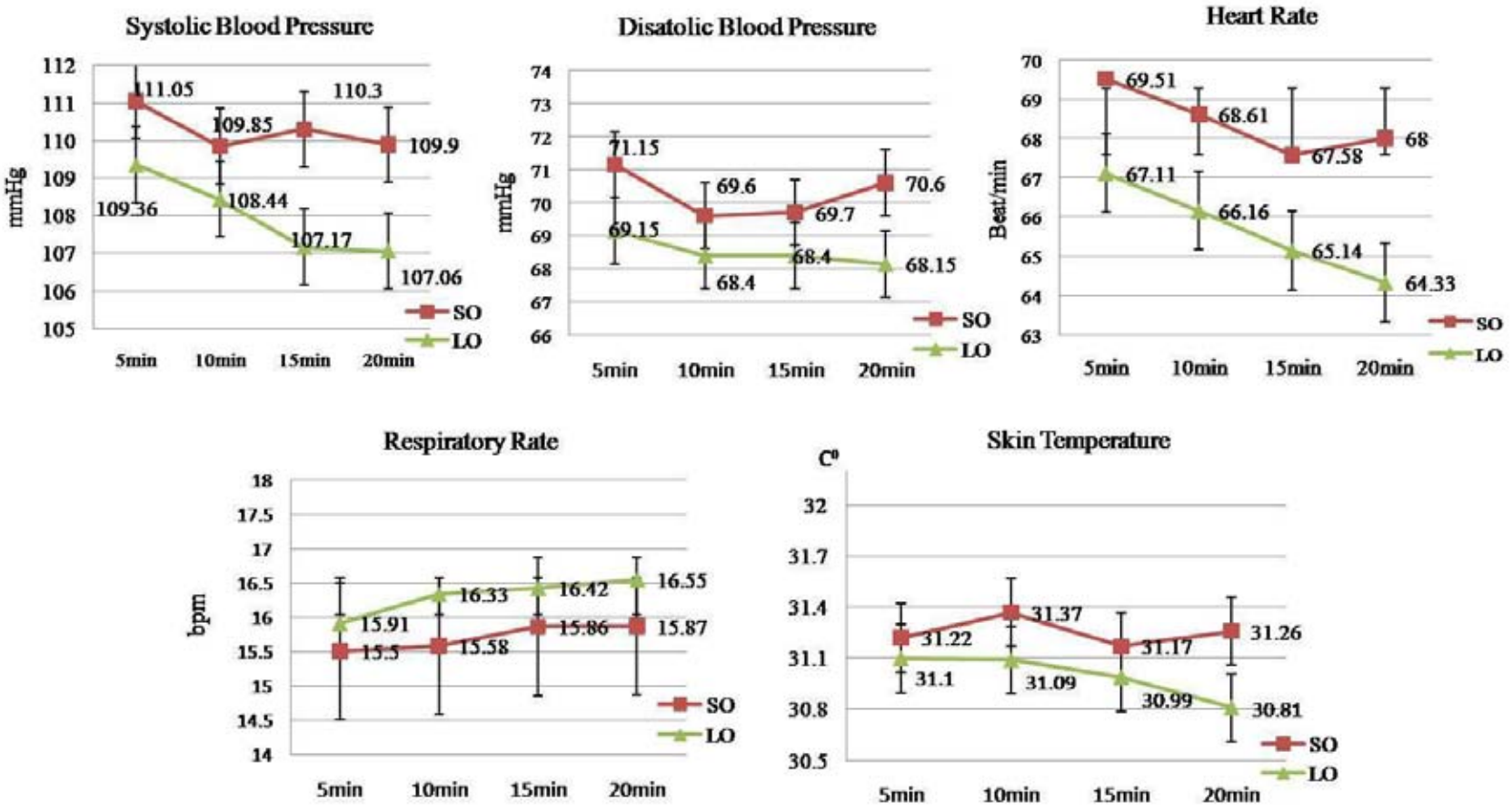


Figure 9 The schematic diagrams of the comparison on autonomic nervous system change, systolic blood pressure, diastolic blood pressure, heart rate, respiratory rate, skin temperature during inhale sweet almond oil (SO), inhale lavender oil (LO). Line graph shows that volunteers have decreased their blood pressure, heart rate and skin temperature at 5-20 minutes whereas their respiratory rate is increased.

Emotions response

The mean and Standard Deviation (SD) values of mood state response are shown in Table 4. Subjects felt unpleasant during the application of sweet almond oil, with data showing decreased scores in good, active, fresh and relaxed feelings. After a lavender inhalation, subjects reported that they had significant increases in pleasant emotions; active, and relaxed (p-value <0.05). Furthermore, drowsy feelings were significantly decreased (p-value <0.05).

Table 4 Mean and SD values of emotional state change, resting, sweet almond oil and lavender oil inhalations.

Emotion	n	*Rest*		*Sweet almond oil*		*Lavender*		*p*-value R and SO	*p*-value SO and LO
		Mean	SD	Mean	SD	Mean	SD		
Good	20	61.50	11.20	50.05	17.22	73.15	14.78	0.010*	0.001*
Bad	20	15.50	12.77	23.55	18.57	15.12	18.71	0.046*	0.085
Active	20	50.80	16.18	44.05	13.83	64.20	13.66	0.104	0.001*
Drowsy	20	26.55	19.47	40.90	24.47	30.05	20.22	0.003*	0.047*
Fresh	20	53.45	12.68	43.35	11.12	59.40	18.18	0.004*	0.001*
Relax	20	59.15	20.97	51.55	19.26	73.65	21.41	0.243	0.004*
Stress	20	12.55	8.75	16.25	12.38	16.45	11.35	0.086	0.948
Uncomfortable	20	16.85	13.45	24.00	16.94	18.70	15.22	0.627	0.206
Romantic	20	28.78	17.43	31.35	22.86	40.55	24.38	0.709	0.151
Frustrated	20	12.51	10.15	16.40	14.77	16.55	18.38	0.094	0.976
Calm	20	62.00	18.96	54.85	19.98	61.60	20.18	0.112	0.276
Disgust	20	8.60	7.60	12.35	11.60	10.85	14.11	0.092	0.712

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), lavender oil (LO)

EEG data

The mean and Standard Deviation (SD) of power value is presented in Table 5. There were noticeable changes of band power in theta and alpha waves that significantly increased during the lavender inhalation over all brains areas (*p*-value <0.05). However, band powers in beta waves were not significantly different. The present study examined changes in the anterior, posterior alpha asymmetry left and right side response to sweet almond oil and lavender. There was no significant asymmetry (*p*-value > 0.05). The topographic map shows obviously more scattering

power in alpha brain, particularly in bilateral temporal and central area after smelling lavender compared with resting and sweet almond oil as shown in Figure 10.

Table 5 Mean and SD power values in eyes closed state, sweet almond oil and lavender oil inhalations.

Area	Theta Power (μV^2)						*p*-value R and SO	*p*-value SO and LO
	Eye close		*Sweet almond oil*		*Lavender*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	1.91	0.18	1.54	0.18	2.16	0.25	0.590	0.001*
right anterior	2.00	0.18	1.62	0.18	2.27	0.27	0.090	0.001*
Center	2.68	0.24	2.10	0.24	3.05	0.36	0.030*	0.006*
left posterior	1.13	0.14	1.08	0.16	1.34	0.21	0.550	0.002*
right posterior	1.15	0.15	1.10	0.18	1.38	0.23	0.025*	0.025*
Area	**Alpha 1 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and LO**
	Eye close		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	3.44	1..18	3.71	1.60	6.94	2.36	0.218	0.001*
right anterior	4.02	1.61	4.38	2.07	7.70	2.66	0.218	0.001*
Center	4.78	1.46	4.83	1.96	9.40	3.07	0.156	0.001*
left posterior	4.16	2.08	4.56	2.57	6.86	2.77	0.218	0.001*
right posterior	4.29	1.94	4.46	2.21	8.79	3.68	0.001*	0.001*
Area	**Alpha 2 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and LO**
	Eye close		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	1.51	0.22	1.43	0.21	2.09	0.30	0.911	0.011*
right anterior	1.63	0.26	1.50	0.24	2.23	0.35	0.575	0.006*
Center	2.28	0.33	1.96	0.30	3.09	0.42	0.179	0.003*
left posterior	2.37	0.54	2.31	0.53	3.41	0.68	0.823	0.008*
right posterior	2.76	0.55	2.51	0.54	4.10	0.80	0.002*	0.002*
Area	**Beta Power (μV^2)**						***p*-value R and SO**	***p*-value SO and LO**
	Eye close		***Sweet almond oil***		***Lavender***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	0.31	0.03	0.35	0.03	0.33	0.03	0.167	0.351
right anterior	0.32	0.04	0.36	0.04	0.35	0.04	0.156	0.433
Center	0.36	0.05	0.41	0.05	0.41	0.05	0.086	0.627
left posterior	0.31	0.04	0.37	0.06	0.36	0.05	0.156	0.852
right posterior	0.31	0.03	0.36	0.04	0.37	0.05	0.794	0.794

*Significant difference, *p*-value < 0.05 eye close (EC), sweet almond oil (SO), lavender oil (LO)

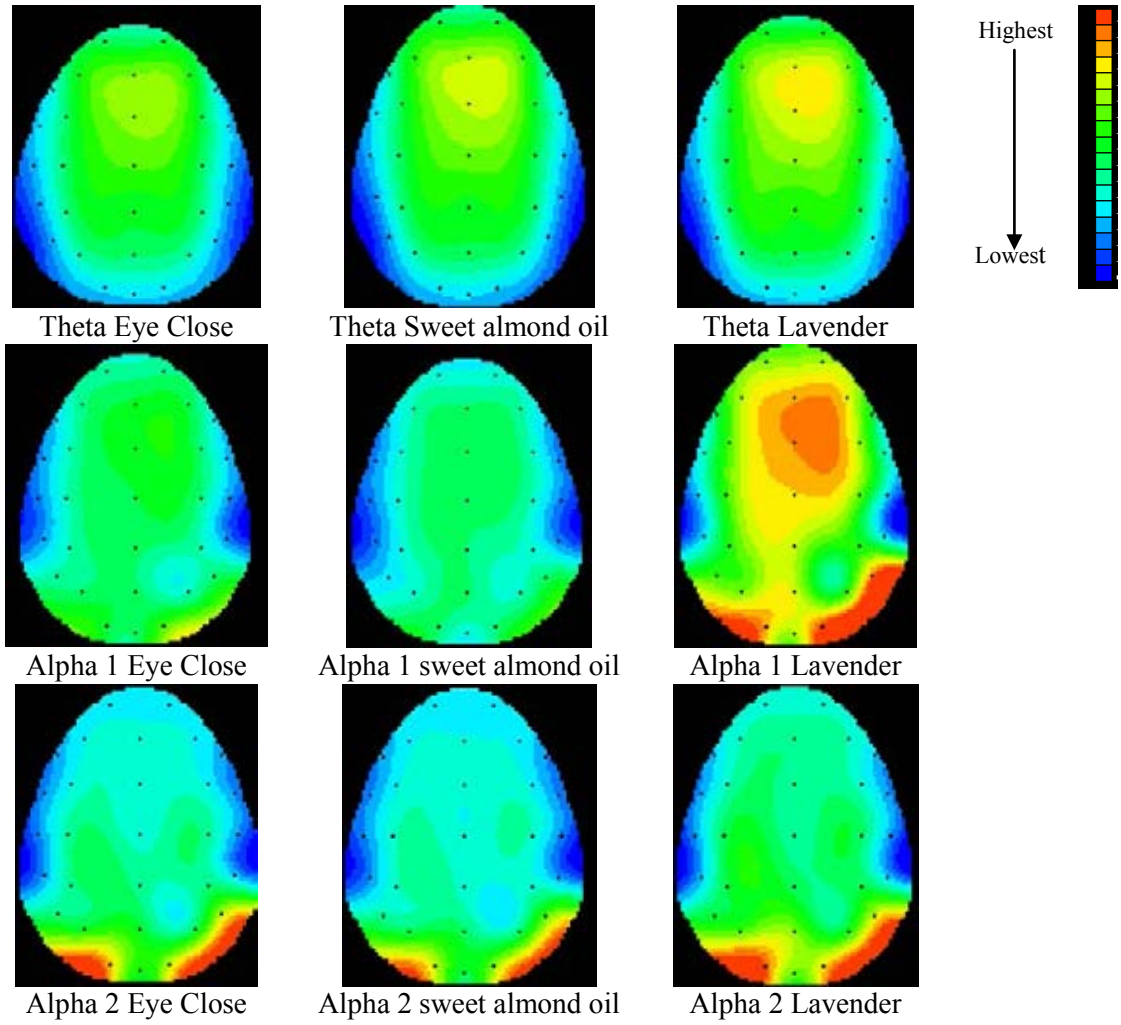


Figure 10 The schematic diagram of the grand average on Theta, Alpha 1, Alpha 2 topographical brain mapping in eye close, inhale sweet almond oil and inhale lavender.

The color bar represents the amount of energy in μV unit; maximum in red scale and minimum in blue scale. Topography, which shows colors of Theta power occurred in volunteer with lavender oil inhalation, of frontal and center brain areas demonstrated a color change into dark yellow whereas color of posterior area is changed into green. Likewise, Alpha power occurred during sweet almond oil inhalation is represented an increase of green color while an increase of reddish yellow color is seen during lavender oil inhalation.

The Correlation between physiological parameters and emotional

In table 6 revealed the change correlation between physiological parameters, including autonomic parameters, power brain wave and emotion by using Spearman rank-order correlation coefficient for statistical analyses. Calm emotion showed invert correlation with diastolic blood pressure (r = -0.521, p = 0.032) while drowsy showed correlation in skin temperature (r = 0.567, p = 0.044). The correlation between brain power and emotions revealed that the change in calm emotion was correlated to anterior alpha 1 power in both sides of the brain (right: r = 0.620, p = 0.043). Otherwise, no significantly difference between physiological and emotional parameters.

Table 6 Correlation among changes of three parameters: autonomic parameters, brain wave power and emotional state after lavender oil inhalations.

Emotion	**ANS**	**Brain wave**		
		Alpha 1	**Alpha 2**	**Beta**
Calm	Diastolic BP (r = -0.521*) p = 0.032	LA (r = 0.520*) p = 0.030 RA (r = 0.490*) p = 0.040		
Drowsy	Skin Temperature (r = 0.567) p = 0.044			
Fresh				Cen (r = 0.62) p = 0.043

*Significant correlation, *p*-value < 0.05, r (Correlation coefficient), Diastolic BP (Diastolic blood pressure), LA (left anterior), RA (right anterior), Cen (center)

Rosemary oil

Rosemary oil components

The composition of rosemary oil was analyzed by gas chromatography/ mass spectrometry (GC/MS) equipped with Finnigan DSQ MS detector, Thermo Finnigan model Trace GC Ultra. The identification of the oil's constituents was performed using their mass spectra and retention times by NIST05 MS library; the percentage compositions also were computed from GC peak area. The result revealed that rosemary oil consisted mainly of 19.41% α-pinene, 20.08% 1, 8-cineole and 21.25% camphor (Appendix J).

The general characteristics of the participants

A total of 20 healthy (10 males and 10 females) subjects aged between 18 to 28 years (mean age 21 ± 2.97 years) with a body mass index ranged 18-23 kg/m^2 (mean BMI 20.69 ± 1.69) were enrolled in this study. As a number of studies have indicated that there is a different activity in the left-handed and right-handed subjects during olfactory tasks, only right-hander was tested. Handedness was tested using Edinburgh Handedness Inventory scale. The subjects were then screened for a normal sense of smell by the n-butyl alcohol method test (mean score 9.65 ± 0.96) shown in Table 7.

Table 7 Demographic data for the rosemary inhaling participants.

Parameters	Number	Minimum	Maximum	Mean	SD
Age	20	18	28	21.00	2.97
Height(cm)	20	154	179	168.25	5.12
Weight (kg)	20	45	72	58.57	6.15
Body Mass Index (kg/m^2)	20	18.21	23.23	20.69	1.69
Smell test	20	9	11	9.65	0.96

Autonomic Nervous System Parameters

The mean and Standard Deviation (SD) values of the ANS parameters in the experiment are presented in Table 8. The data on various ANS parameters were compared during resting, during inhalation of sweet almond oil and during inhalation of rosemary oil. Subjects had significantly decreased their heart rate (p-value = 0.043) during the sweet almond oil inhalation compared with those of resting. However, the blood pressure, heart rate and respiratory rate showed a significant increase upon exposure to rosemary oil. The skin temperature, on the contrary, decreased significantly shown in Figure 11.

Table 8 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and rosemary oil inhalations.

Minute	Systolic Blood Pressure (mmHg)						*p*-value R and SO	*p*-value SO and RO
	Rest		*Sweet almond oil*		*rosemary oil*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	106.10	8.72	106.80	9.27	107.35	8.92		
10	104.70	8.36	106.05	9.06	107.70	9.13		
15			105.10	7.92	108.40	9.02		
20			104.90	8.47	109.95	8.67		
Average	105.40	8.54	105.71	8.57	108.31	8.88	0.624	0.000*
Minute	**Diastolic Blood Pressure (mmHg)**						***p*-value R and SO**	***p*-value SO and RO**
	Rest		*Sweet almond oil*		*rosemary oil*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	64.34	10.72	62.60	6.08	68.50	8.12		
10	64.18	11.36	64.75	5.62	68.25	9.72		
15			64.65	5.95	71.53	6.63		
20			63.75	5.72	72.40	7.08		
Average	64.26	11.04	63.93	5.80	70.17	7.97	0.870	0.000*
Minute	**Heart rate(beat/min)**						***p*-value R and SO**	***p*-value SO and RO**
	Rest		*Sweet almond oil*		*rosemary oil*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	72.25	11.27	69.83	9.70	71.76	11.13		
10	71.69	12.31	68.61	9.95	71.52	9.55		
15			69.53	9.49	72.66	10.74		
20			69.77	9.63	73.09	10.11		
Average	71.97	11.19	69.43	9.52	72.25	10.22	0.043*	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Rosemary oil (RO)

Table 8 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and rosemary oil inhalations (Continue).

Minute	Respiratory rate (bpm)						*p*-value R and SO	*p*-value SO and RO
	Rest		*Sweet almond oil*		*rosemary oil*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	16.02	1.75	15.04	2.35	15.97	2.98		
10	15.94	2.17	15.65	2.86	16.75	2.64		
15			16.19	2.96	16.61	2.42		
20			16.03	1.95	16.99	2.64		
Average	15.98	1.96	15.72	2.55	16.58	2.65	0.426	0.000*
Minute	**Skin temperature (°C)**						***p*-value R and SO**	***p*-value SO and RO**
	Rest		*Sweet almond oil*		*rosemary oil*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	31.95	1.90	32.12	1.93	31.84	1.97		
10	32.29	1.78	32.25	2.04	31.77	1.99		
15			32.33	2.08	31.82	1.89		
20			32.27	2.00	31.74	1.82		
Average	32.12	1.84	32.24	1.97	31.79	1.88	0.507	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Rosemary oil (RO)

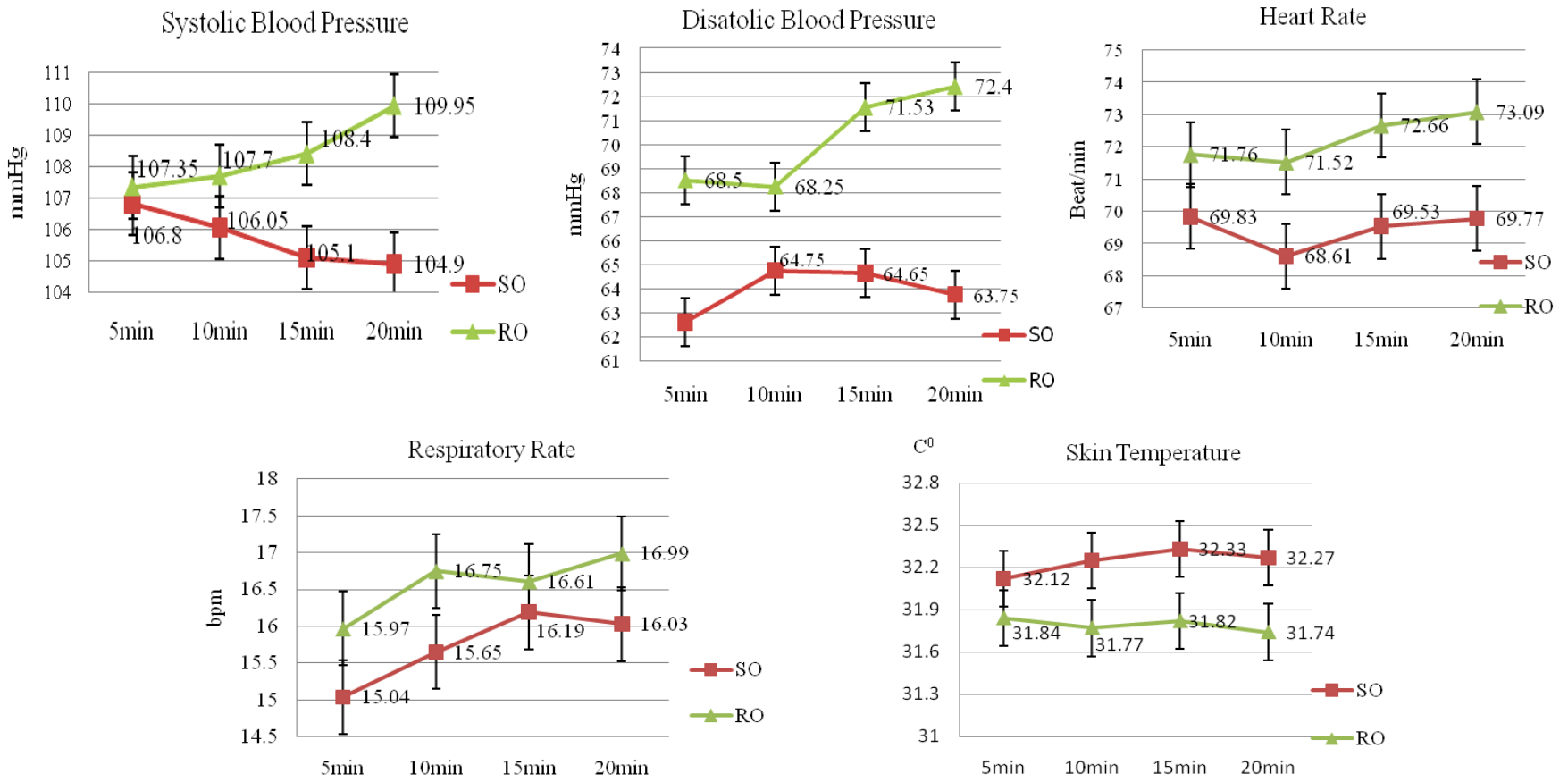


Figure 11 The schematic diagram of the comparison on autonomic nervous system changes, systolic blood pressure, diastolic blood pressure, heart rate, respiratory rate, skin temperature during inhale sweet almond oil (SO), inhale rosemary oil (RO).Line graph shows that volunteers have increased on their blood pressure, heart rate and respiratory rate since at 5-20 minutes whereas their skin temperature is decreased.

Emotions response

The mean and Standard Deviation (SD) values of the mood state responses are shown in table 9. In the second trial, the subjects became significantly less active after they were administered sweet almond oil, as compared to baseline condition (at rest). In the third trial, exposure to rosemary oil increased positive emotions including the feeling of well-being, active and fresh (p-value < 0.05). Furthermore, drowsiness was significantly reduced when compared between rosemary oil and sweet almond oil inhalations (p-value < 0.05).

Table 9 Mean and SD values of emotional state change, resting, sweet almond oil and Rosemary oil inhalations.

Emotion	n	*Rest*		*Sweet almond oil*		*Rosemary*		*p*-value R and SO	*p*-value SO and RO
		Mean	SD	Mean	SD	Mean	SD		
Good	20	66.30	16.73	66.85	18.94	76.38	16.90	0.806	0.005
Bad	20	16.05	11.90	14.85	13.31	12.00	14.58	0.594	0.551
Active	20	52.05	14.28	41.10	20.83	64.57	23.29	0.074	0.000*
Drowsy	20	40.00	22.62	51.85	10.78	39.10	24.94	0.028*	0.032*
Fresh	20	54.40	21.42	48.35	20.65	69.31	22.74	0.263	0.000*
Relax	20	65.70	19.27	60.70	17.06	65.78	18.60	0.304	0.074
Stress	20	19.10	7.55	15.80	8.54	11.63	3.17	0.364	0.294
Uncomfortable	20	25.90	11.93	18.65	13.31	13.21	10.98	0.103	0.098
Romantic	20	28.15	13.81	40.80	21.56	48.42	13.66	0.149	0.785
Frustrated	20	16.50	5.81	13.84	1.95	11.42	3.57	0.350	0.298
Calm	20	65.40	15.43	61.73	16.80	63.21	13.73	0.298	0.598
Disgust	20	10.90	8.50	9.31	6.36	6.59	3.51	0.695	0.076

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Rosemary oil (RO)

EEG data

The mean and Standard Deviation (SD) values of power values are presented in Table 10. In rosemary session, the band power of alpha1 in the left and right anterior and right posterior regions showed a significant decrease (p-value < 0.05). There were noticeable changes of band power in alpha2 waves that significantly decreased during the rosemary inhalation in all brains areas (p-value < 0.05). Conversely, the band power of beta wave in the left and right anterior was significantly increased. No significant change was observed in the case of theta power

(p-value > 0.05). In Figure 12, the topographic map shows after smelling rosemary compared with resting and sweet almond oil obviously less spreading power in alpha brain, particularly in bilateral temporal and central area. In contrast, beta brain power increased in frontal area.

Table 10 Mean and SD power values in eye closed state, sweet almond oil and rosemary oil inhalations.

Area	Theta Power (μV^2)						*p*-value R and SO	*p*-value SO and RO
	Eye close		*Sweet almond oil*		*Rosemary*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	1.94	0.23	2.11	0.24	1.95	0.25	0.089	0.345
right anterior	2.30	0.25	2.19	0.26	2.01	0.27	0.100	0.297
Center	2.78	0.33	2.99	0.34	2.80	0.39	0.100	0.362
left posterior	1.40	0.25	1.53	0.28	1.43	0.29	0.070	0.176
right posterior	1.34	0.19	1.46	0.19	1.35	0.18	0.160	0.234
Area	**Alpha 1 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and RO**
	Eye close		*Sweet almond oil*		*Rosemary*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	5.25	1.08	6.35	1.20	5.42	1.21	0.11	0.043*
right anterior	5.39	1.05	6.58	1.22	5.57	1.24	0.09	0.031*
Center	8.56	1.74	10.15	1.91	8.74	2.03	0.11	0.065
left posterior	6.87	1.81	7.80	2.18	6.01	1.67	0.36	0.074
right posterior	8.03	2.16	9.59	2.35	6.88	1.65	0.19	0.033*
Area	**Alpha 2 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and RO**
	Eye close		*Sweet almond oil*		*Rosemary*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	2.71	0.56	2.72	0.54	2.20	0.41	0.94	0.015*
right anterior	2.68	0.51	2.63	0.48	2.14	0.37	0.77	0.018*
Center	4.32	0.82	4.60	0.91	3.62	0.76	0.497	0.031*
left posterior	4.77	1.10	5.30	1.34	3.57	0.85	0.351	0.031*
right posterior	5.30	1.13	5.82	1.35	3.98	0.87	0.348	0.023*
Area	**Beta Power (μV^2)**						***p*-value R and SO**	***p*-value SO and RO**
	Eye close		*Sweet almond oil*		*Rosemary*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	0.29	0.03	0.31	0.04	0.36	0.03	0.610	0.025*
right anterior	0.31	0.04	0.31	0.05	0.37	0.04	0.778	0.045*
Center	0.38	0.05	0.39	0.06	0.42	0.06	0.535	0.264
left posterior	0.35	0.05	0.37	0.06	0.37	0.06	0.567	0.862
right posterior	0.33	0.04	0.38	0.06	0.36	0.06	0.191	0.655

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Rosemary oil (RO)

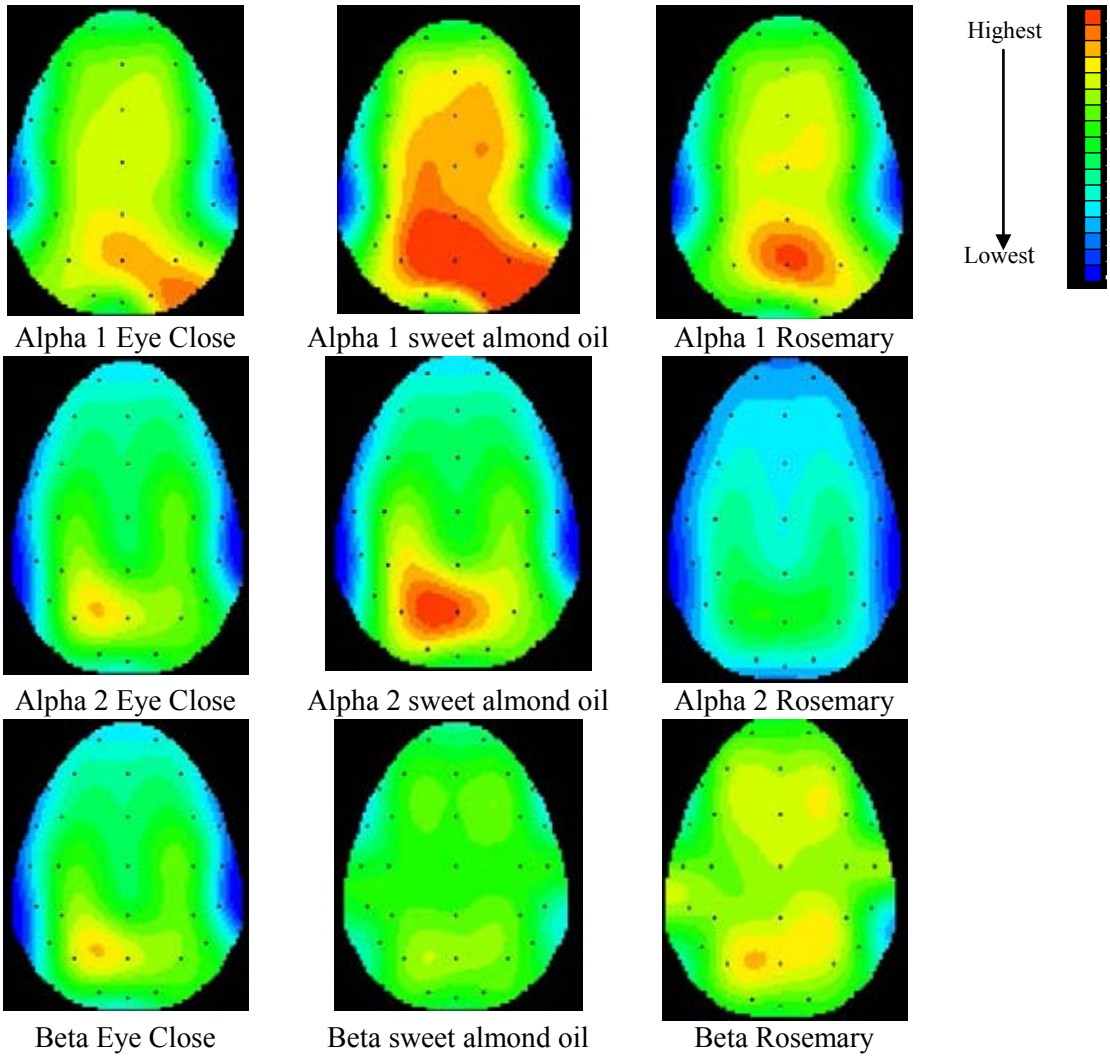


Figure12 The schematic diagram of the grand average of Alpha 1, Alpha 2, Beta topographical brain mapping in eye close, inhale sweet almond oil and inhale rosemary.

The color bar represents the amount of energy in μV unit; maximum in red scale and minimum in blue scale. Topography color of Alpha power shows faded orange color whereas darker yellow is seen during sweet almond oil inhalation. During rosemary oil inhalation, color is also faded. In contrast, Beta power is increased by color changing from green into yellowish orange.

The Correlation between physiological parameters and emotional

Table 11 revealed the correlation change between physiological parameters, including autonomic parameters, power brain wave and emotion by using Spearman rank-order correlation coefficient for statistical analyses. Active emotion showed a correlation with systolic blood pressure (r = 0.535, p = 0.045). The correlation among parameters revealed that the change in relaxing was correlated to left anterior alpha1 power in both sides of the brain (right: r = 0.560, p = 0.010), (left: r = 0.56, p = 0.010) and center (r = 0.636, p = 0.003). Furthermore, relaxation was correlated to right posterior alpha 2 power (r = 0.476, p = 0.034). Fresh emotion was correlated with beta power in center (r = 0.473, p = 0.035) reverse alpha 1 power in both anterior sides (Left r = -0.472, p = 0.036), (right r = -0.440, p = 0.042). Drowsy inverse correlated with beta power in right anterior (r = -0.589, p = 0.010) and center (r = -0.530, p = 0.040); otherwise there was no significant correlation between physiological and emotional parameters.

Table 11 Correlation among changes of three parameters: autonomic parameters, brain wave power and emotional state after rosemary oil inhalations.

Emotion	**ANS**	**Brain wave**		
		Alpha 1	**Alpha 2**	**Beta**
Active	Systolic BP (r = 0.535*) p = 0.037			
Drowsy				RA (r = -0.589*) p = 0.010 Cen (r = -0.530*) p = 0.040
Fresh		LA (r = -0.472*) p = 0.040 RA (r = -0.440*) p = 0.040		Cen (r = 0.473*) p = 0.040
Relax		LA (r=0.564*) p = 0.010 RA (r = 0.560*) p = 0.010 Cen (r = 0.636*) p = 0.000	RP (r = 0.476*) p = 0.034	

*Significant correlation, *p*-value < 0.05, r (Correlation coefficient), Systolic BP (Systolic blood pressure), LA (left anterior), RA (right anterior), cen (center), RP (right posterior)

Jasmine oil

Jasmine oil components

The composition of jasmine oil was analyzed by gas chromatography/mass spectrometry (GC/MS) equipped with Finnigan DSQ MS detector, Thermo Finnigan model Trace GC Ultra. The oil's constituents were identified by their mass spectra and retention times from NIST05 MS library; the percentage compositions also were computed from GC peak area. The result revealed that jasmine oil mainly of 26.09% Benzyl acetate, 11.02% Beta-Linalool, 9.65% Benzyl propionate (Appendix J).

General characteristics of the participants

A total of 20 healthy (10 males and 10 females) subjects aged between 18 to 32 years (mean age 22.70 ±4.27 years) with a body mass index ranged 18-25 kg/m^2 (mean BMI 21.33 ± 2.10) were enrolled in this study. As a number of studies have indicated that there is a different activity between the left-handed and right-handed subjects during olfactory tasks, only right-hander was tested. Handedness was tested using Edinburgh Handedness Inventory scale. The subjects were then screened for a normal sense of smell by the n-butyl alcohol method test (mean score 9.60 ± 0.86). A summary of the demographic data of the participants is presented in Table 12.

Table 12 Demographic data for the jasmine inhaling participants.

Parameters	Number	Minimum	Maximum	Mean	SD
Age	20	18	32	22.70	4.27
Height(cm)	20	154	179	168.25	5.12
Weight (kg)	20	45	72	58.57	6.15
Body Mass Index (kg/m^2)	20	18.21	24.97	21.33	2.10
Smell test	20	8	11	9.60	0.86

Autonomic Nervous System Parameters

The mean and Standard Deviation (SD) values of the ANS parameters in the experiment are presented in Table 13 and Figure13. The data on various ANS parameters were compared during resting, inhalation of sweet almond oil and jasmine oil. Subjects had significantly decreased their heart rate (p-value = 0.001) during the sweet almond oil inhalation compared to those of resting. However, blood pressure, heart rate and respiratory rate showed a significant increase upon exposure to jasmine oil. The skin temperature, on the contrary, showed not significantly change.

Table 13 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and jasmine oil inhalations.

Minute	Systolic Blood Pressure (mmHg)						*p*-value R and SO	*p*-value SO and JO
	Rest		*Sweet almond oil*		*Jasmine*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	104.60	9.52	104.70	8.95	106.20	9.49		
10	104.20	9.28	104.25	8.75	107.15	9.14		
15			104.40	9.11	107.85	8.79		
20			103.80	9.37	108.50	9.19		
Average	104.40	9.40	104.28	8.88	107.42	9.02	0.831	0.000*
Minute	**Diastolic Blood Pressure (mmHg)**						***p*-value R and SO**	***p*-value SO and JO**
	Rest		***Sweet almond oil***		***Jasmine***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	64.15	9.71	61.35	7.22	65.30	6.65		
10	62.35	10.11	61.45	8.27	66.35	9.21		
15			60.70	8.38	67.55	9.69		
20			61.50	7.95	68.75	8.07		
Average	63.25	9.64	61.25	7.82	66.98	8.42	0.516	0.000*
Minute	**Heart rate(beat/min)**						***p*-value R and SO**	***p*-value SO and JO**
	Rest		***Sweet almond oil***		***Jasmine***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	72.12	10.55	70.55	9.61	73.04	9.38		
10	69.84	10.51	69.58	9.09	74.18	9.48		
15			69.75	9.18	74.84	9.24		
20			69.57	9.42	75.37	8.96		
Average	70.98	10.53	69.86	9.16	74.35	9.13	0.001*	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), jasmine oil (JO)

Table 13 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and jasmine oil inhalations (Continue).

Minute	Respiratory rate (bpm)						*p*-value R and SO	*p*-value SO and JO
	Rest		*Sweet almond oil*		*Jasmine*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	16.61	2.75	15.40	3.33	17.53	4.48		
10	15.13	2.96	15.77	2.40	17.67	3.33		
15			16.13	2.68	17.12	2.46		
20			16.32	3.99	17.42	2.02		
Average	15.87	2.72	15.90	3.12	17.43	3.16	0.824	0.000*
Minute	**Skin temperature (°C)**						***p*-value R and SO**	***p*-value SO and JO**
	Rest		*Sweet almond oil*		*Jasmine*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	31.94	2.08	31.97	1.95	32.10	1.81		
10	31.72	2.02	32.09	1.85	32.10	1.89		
15			32.16	1.81	31.99	1.91		
20			32.23	1.86	31.92	1.89		
Average	31.83	2.05	32.11	1.83	32.02	1.84	0.341	0.116

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), jasmine oil (JO)

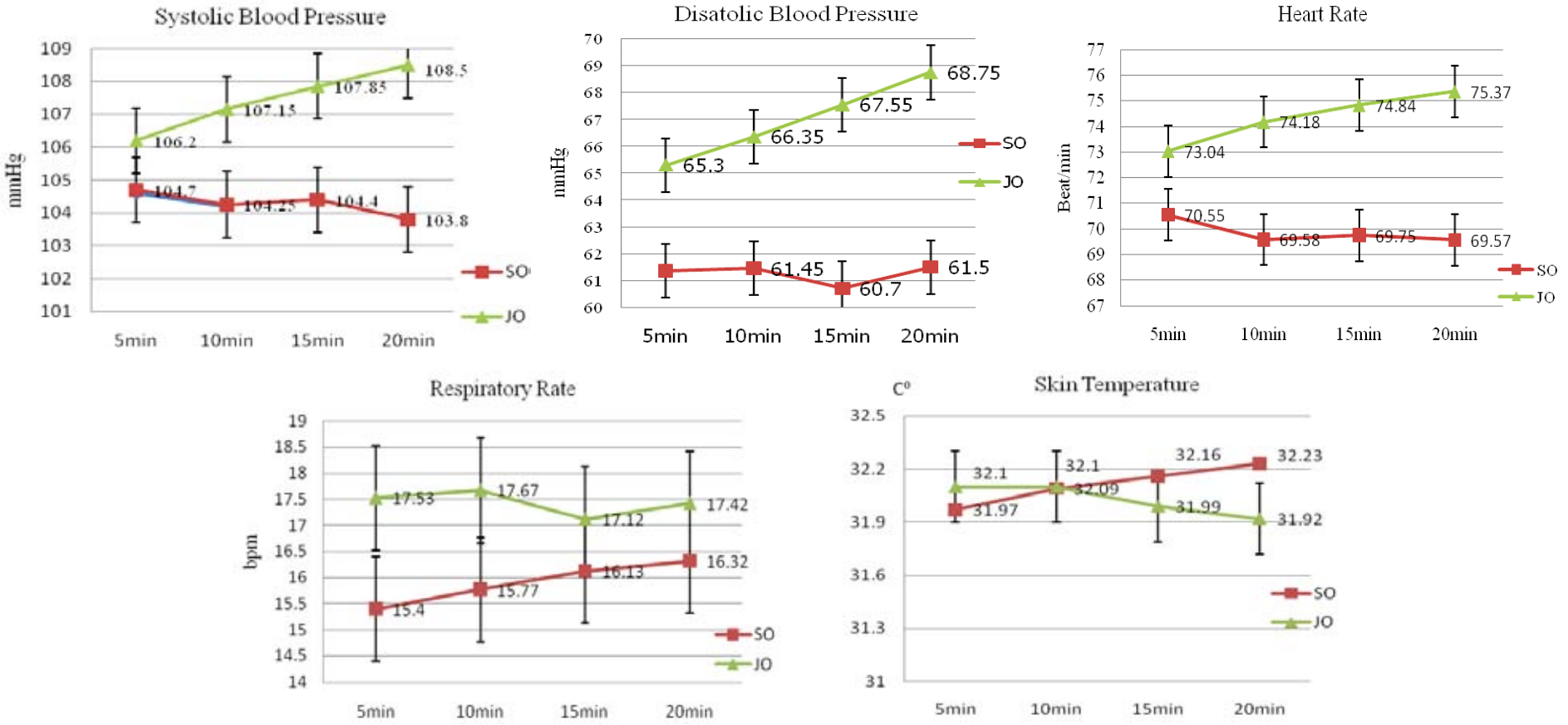


Figure13 The schematic diagram of the comparison autonomic nervous system change, systolic blood pressure, diastolic blood pressure, heart rate, respiratory rate, skin temperature during inhale sweet almond oil (SO), inhale jasmine oil (JO). Graph depicts that volunteers have increased on their blood pressure, heart rate and respiratory rate since at 5-20 minutes, except their skin temperature during both conditions is similar.

Emotions response

The mean and Standard Deviation (SD) values of the mood state responses are shown in table14. In the second trial, the subjects became significantly less fresh after inhaling sweet almond oil, as compared to baseline condition (at rest). In the third trial, exposure to jasmine oil increased positive emotions including the feeling of well-being, active fresh and romantic (p-value < 0.05). Furthermore, negatively emotion such as drowsy, uncomfortable and disgust feeling were significantly reduced (p-value < 0.05).

Table 14 Mean and SD values of emotional state change, resting, sweet almond oil and jasmine oil inhalations.

Emotion	n	*Rest*		*Sweet almond oil*		*Jasmine*		*p*-value R and SO	*p*-value SO and JO
		Mean	SD	Mean	SD	Mean	SD		
Good	20	55.90	14.46	54.30	18.18	78.10	12.74	0.620	0.000*
Bad	20	16.60	4.29	23.50	8.52	13.75	4.30	0.170	0.080
Active	20	50.40	13.78	37.10	19.02	53.40	20.56	0.062	0.014*
Drowsy	20	33.20	19.41	40.85	17.35	30.10	16.81	0.121	0.042*
Fresh	20	55.20	14.96	41.85	17.35	60.80	17.84	0.015*	0.002*
Relax	20	56.75	17.28	51.00	19.48	68.15	14.19	0.328	0.002*
Stress	20	21.35	7.82	12.70	8.42	10.50	6.90	0.067	0.208
Uncomfortable	20	19.00	12.57	19.15	11.94	12.80	7.70	0.959	0.005*
Romantic	20	34.40	19.93	30.55	22.07	49.05	22.76	0.582	0.007*
Frustrated	20	14.40	13.76	15.00	10.33	13.00	14.45	0.861	0.643
Calm	20	58.65	20.76	53.65	20.09	59.30	19.37	0.435	0.311
Disgust	20	7.68	4.42	17.00	6.65	8.35	3.29	0.311	0.027*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Jasmine oil (JO)

EEG data

The mean and Standard Deviation (SD) of power value are shown in Table 15. Referring to jasmine session, the band power of beta in the left and right anterior center and left regions showed a significant increase (p-value < 0.05). However, theta and alpha band powers decreased with no significant statistic change (p-value > 0.05). In Figure 14, the topographic map shows after inhaling jasmine compared with resting and sweet almond oil inhalation. The beta wave power increased obviously in bilateral frontal and posterior.

Table 15 Mean and SD power values in eye closed state, sweet almond oil and jasmine oil inhalations.

Area	Theta Power (μV^2)						*p*-value R and SO	*p*-value SO and JO
	Eye close		*Sweet almond oil*		*Jasmine*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	3.36	0.43	3.62	0.49	3.56	0.46	0.274	0.596
right anterior	3.59	0.44	3.71	0.49	3.79	0.46	0.397	0.365
Center	5.71	0.84	5.44	0.88	5.49	0.78	0.402	0.857
left posterior	3.11	0.62	3.30	0.86	3.61	0.58	0.513	0.432
right posterior	3.40	0.55	3.56	0.73	3.76	0.51	0.488	0.589
Area	**Alpha 1 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and JO**
	Eye close		***Sweet almond oil***		***Jasmine***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	8.86	2.59	9.02	2.50	7.74	1.54	0.670	0.330
right anterior	9.25	2.51	9.51	2.44	8.30	1.57	0.537	0.344
Center	14.06	3.88	14.03	3.71	12.15	2.39	0.967	0.314
left posterior	10.05	2.88	10.62	3.14	9.21	2.311	0.478	0.336
right posterior	10.50	2.77	10.89	2.91	9.28	1.93	0.539	0.329
Area	**Alpha 2 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and JO**
	Eye close		***Sweet almond oil***		***Jasmine***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	1.89	0.25	2.09	0.27	1.97	0.22	0.063	0.473
right anterior	1.98	0.26	2.21	0.29	2.10	0.25	0.053	0.526
Center	3.11	0.45	3.30	0.46	3.36	0.53	0.207	0.818
left posterior	3.11	0.56	3.31	0.56	3.61	0.70	0.313	0.432
right posterior	3.40	0.58	3.57	0.56	3.76	0.73	0.488	0.589
Area	**Beta Power (μV^2)**						***p*-value R and SO**	***p*-value SO and JO**
	Eye close		***Sweet almond oil***		***Jasmine***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	0.33	0.04	0.34	0.04	0.43	0.04	0.078	0.009*
right anterior	0.34	0.04	0.36	0.04	0.44	0.04	0.072	0.009*
Center	0.44	0.05	0.46	0.05	0.53	0.05	0.071	0.039*
left posterior	0.32	0.04	0.34	0.05	0.45	0.04	0.154	0.017*
right posterior	0.36	0.04	0.39	0.05	0.45	0.04	0.118	0.075

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Jasmine oil (JO)

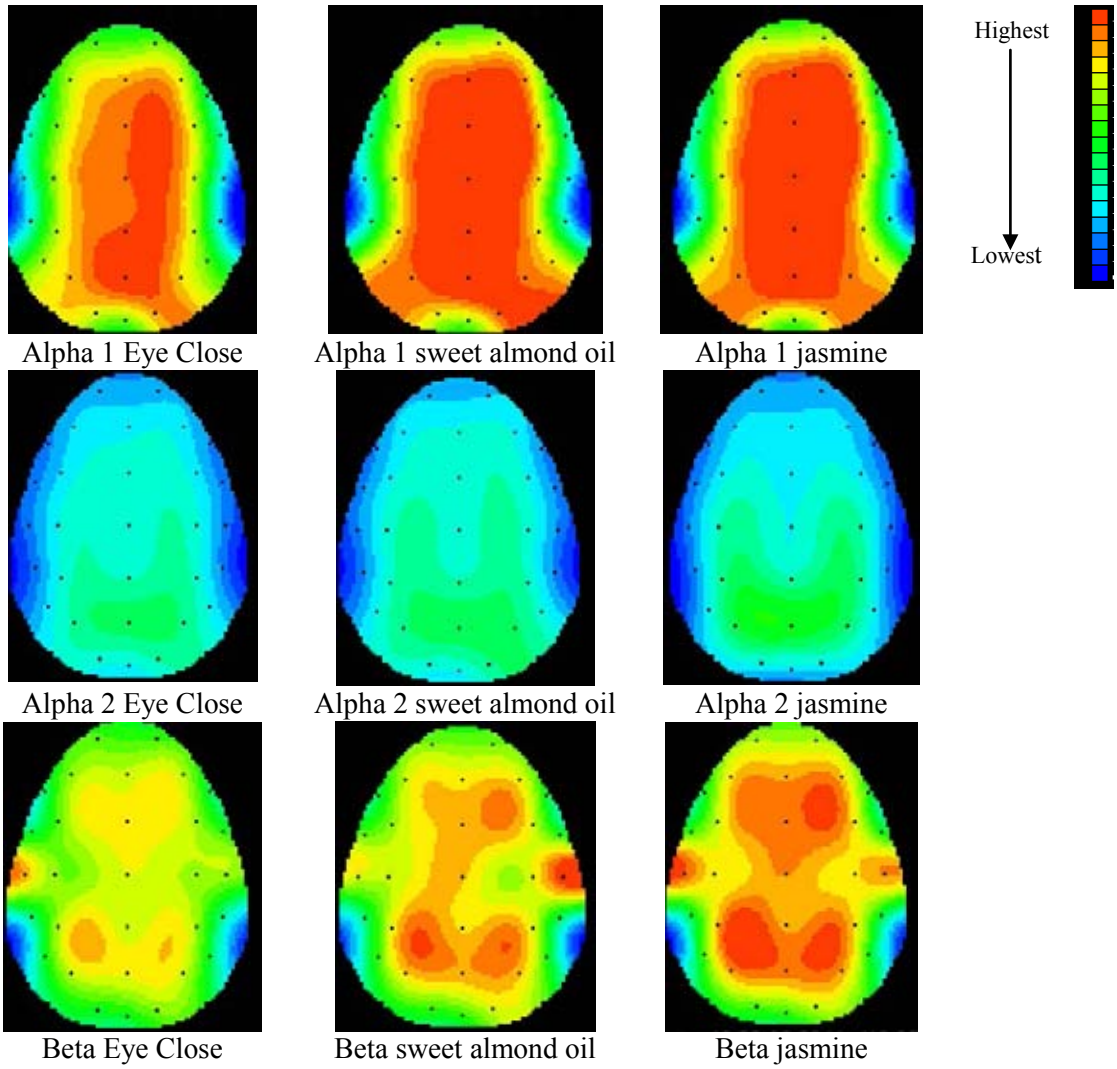


Figure14 The schematic diagram of the grand average of Alpha 1, Alpha 2, Beta topographical brain mapping in eye close, inhale sweet almond oil and inhale jasmine.

The color bar represents the amount of energy in μV unit; maximum in red scale and minimum in blue scale. Topography range of both Alpha power 1 and 2 during sweet almond oil inhalation shows no change in color, as well as during jasmine oil inhalation. However, Beta power occurred during jasmine oil inhalation shows an increase by color changing from dark yellow to orange.

Correlation analysis

Table16 revealed the correlation change between physiological parameters, including autonomic parameters, power brain wave and emotion by using Spearman rank-order correlation coefficient for statistical analyses. Active and fresh emotion showed correlation with heart rate (r = 0.573, p = 0.035, r = 0.524, p = 0.042) respectively, but drowsy emotion showed invert (r = -0.536, p = 0.015) . The correlation between brain power and mood state revealed that the change in alpha 1 power correlated to relax (LA r = 0.550, RA r = 0560, Cen r= 0.560) and calm (LP r = 0.490). While inverts correlate frustrate (LA r = -0.590, RA r= -0.510) and disgust (RA r = -0.496, Cen r = -0.458, LP r = -0.480). In alpha 2 power, it correlated to relax (RP r = 0.520) and cal m (RP r = 0.490) . While, inverts correlate stress (RP r = -0.510), uncomfortable (Cen r = -0.510, RP r = -0.540), frustrated (Cen r = -0.500, RP r = -0.670) and disgust (RP r = -0.582). Beta 2 negatively correlated with drowsy (LA r = -0.570, RA r = -0.570) and relax (LA r = -0.570). Otherwise, there was no significantly correlation between physiological and emotional parameters.

Table 16 Correlation among changes of three parameters: autonomic parameters, brain wave power and emotional state after jasmine oil inhalations

Emotion	ANS	Brain wave		
		Alpha 1	**Alpha 2**	**Beta**
Active	HR (r = 0.573*) p = 0.035			
Drowsy	HR (r = -0.536*) p = 0.015			LA (r = -0.570*) p = 0.009 RA (r = -0.570*) p = 0.008
Fresh	HR (r = 0.524*) p = 0.042			
Relax		LA (r = 0.550*) p = 0.048 RA (r = 0.560*) p = 0.010 Cen (r = 0.560*) p = 0.016	RP (r = 0.520*) p = 0.004	LA (r = -0.570*) p = 0.035
Stress			RP (r = -0.510*) p = 0.025	
Uncomfortable			Cen (r = -0.510*) p = 0.025 RP (r = -0.540*) p = 0.031	
Calm		LP (r = 0.490*) p = 0.031	RP (r = 0.440*) p = 0.049	
Disgust		RA (r = -0.496*) p = 0.026 Cen (r = -0.458*) p = 0.042 LP (r = -0.480*) p = 0.032	RP (r = -0.582*) p = 0.007	

*Significant correlation, *p*-value < 0.05, r (Correlation coefficient), HR (Heart rate), LA (left anterior), RA (right anterior), cen (center), RP (right posterior), LP (left posterior)

Citronella oil

Citronella oil components

The oil composition was identified by gas chromatography /mass spectrometry (GC/MS) (Thermo Finnigan model Trace GC Ultra equipped with Finnigan DSQ MS detector, USA). The constituent of the oil was analyzed using their mass spectra and retention times by NIST05 MS library, and the percentage compositions were computed from GC peak area. Citronella oil was consisted of three main kinds on α–citronellal 33.22 %, geraniol 21.12 % and citronellol 13.07% (Appendix J).

General characteristics of the participants

A total of 20 healthy subjects (10 males and 10 females) aged between 18 to 29 years (mean age 21.40 ±2.76 years) with a body mass index ranged 18.5-23 kg/m^2 (mean BMI 20.68 ± 1.89) were enrolled in this study. As a number of studies have indicated that there is a different activity between the left-handed and right-handed subjects during olfactory tasks, only right-hander was tested. Handedness was tested using Edinburgh Handedness Inventory scale. The subjects were then screened for a normal sense of smell by the n-butyl alcohol method test (mean score 9.60 ± 0.74) shown in Table 17.

Table 17 Demographic data for the citronella inhaling participants.

Parameters	Number	Minimum	Maximum	Mean	SD
Age	20	18	29	21.40	2.76
Height(cm)	20	155	176	167.25	4.22
Weight (kg)	20	47	72	56.17	5.15
Body Mass Index (kg/m^2)	20	18.50	23.23	20.68	1.69
Smell test	20	9	11	9.60	0.74

Autonomic Nervous System Parameters

The mean and standard deviation (SD) values of the ANS parameters in the experiment are shown in Table 18 and Figure15. The data on various ANS parameters were compared during resting, inhalation of sweet almond oil and citronella oil. Subjects had significantly decreased heart rate (p-value < 0.05) during the sweet almond oil treatment compared to those of resting. When subjects inhaled the citronella, blood pressures, heart rate and respiratory rate were significantly decreased when compared to sweet almond oil inhalation. The skin temperature, on the contrary, was not significantly changed.

Table 18 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and citronella oil inhalations.

Minute	Systolic Blood Pressure (mmHg)						*p*-value R and SO	*p*-value SO and CO
	Rest		*Sweet almond oil*		*Citronella*			
	Mean	SD	Mean	SD	Mean	SD		
5	105.52	7.64	105.75	8.03	103.60	7.76		
10	105.32	7.58	105.25	7.31	103.75	7.35		
15			104.45	8.21	103.20	8.06		
20			104.85	8.53	102.91	7.78		
Average	105.42	7.61	105.07	7.89	103.36	7.60	0.764	0.000*
Minute	**Diastolic Blood Pressure (mmHg)**						**p-value R and SO**	**p-value SO and CO**
	Rest		**Sweet almond oil**		**Citronella**			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	65.01	8.89	65.10	7.99	63.60	7.99		
10	64.55	8.87	63.80	8.17	63.80	8.09		
15			62.60	7.53	61.70	7.44		
20			63.00	8.10	60.85	7.42		
Average	64.17	8.88	63.62	7.86	62.48	7.70	0.757	0.021
Minute	**Heart rate(beat/min)**						**p-value R and SO**	**p-value SO and CO**
	Rest		**Sweet almond oil**		**Citronella**			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	70.92	13.69	69.00	12.54	67.67	12.65		
10	70.76	13.79	69.01	12.21	66.62	11.82		
15			69.07	11.70	66.44	12.22		
20			69.10	12.38	65.73	12.20		
Average	70.84	13.74	69.04	11.98	66.61	12.01	0.006*	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), citronella oil (CO)

Table 18 Mean and SD values of autonomic nervous system changes under resting and sweet almond oil and citronella oil inhalations (Continue).

Minute	Respiratory rate (bpm)						*p*-value R and SO	*p*-value SO and CO
	Rest		*Sweet almond oil*		*Citronella*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	16.25	2.92	14.95	2.73	14.72	3.00		
10	16.33	2.78	15.04	3.18	14.79	3.20		
15			15.83	3.30	14.73	3.27		
20			16.51	3.73	14.83	3.03		
Average	16.29	2.85	15.58	3.26	14.76	3.07	0.037	0.003*
Minute	**Skin temperature (°C)**						***p*-value R and SO**	***p*-value SO and CO**
	Rest		***Sweet almond oil***		***Citronella***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
5	32.01	2.28	31.45	2.15	31.61	2.27		
10	30.37	2.46	31.57	2.23	31.26	2.80		
15			31.59	2.24	31.52	2.34		
20			31.73	2.22	31.50	2.30		
Average	31.19	2.37	31.58	2.17	31.47	2.40	0.118	0.312

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), citronella oil (CO)

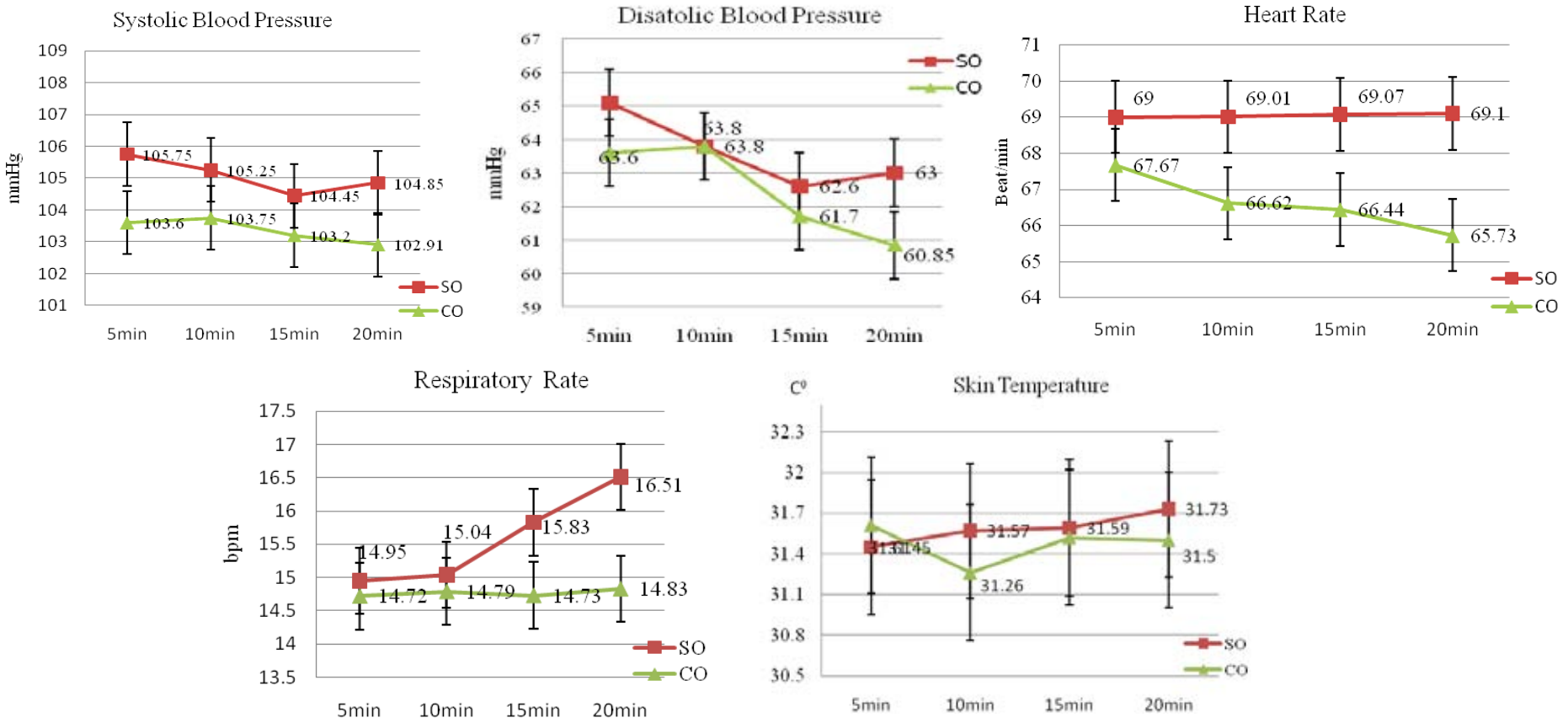


Figure15 The schematic diagram of the comparison autonomic nervous system change systolic blood pressure, diastolic blood pressure, heart rate, r espiratory r ate, skin temperature du ring i nhale s weet a lmond oi l (SO), inhale c itronella o il (CO). Graph s hows t hat vol unteers ha ve decreased in their blood pressure, heart rate and respiratory rate since at 5-20 minutes, except their skin temperature which is similar during both sweet almond oil and citronella oil inhalations.

Emotions response

The mean and Standard Deviation (SD) values of mood state response are shown in Table 19 After a citronella inhalation, subjects felt that they had significant increases in pleasant emotions: good, fresh, relax and calm (p-value < 0.05). No significant change was observed in the case of other mood states (p-value > 0.05).

Table 19 Mean and SD values of emotional state change, resting, sweet almond oil and citronella oil inhalations.

Emotion	n	*Rest*		*Sweet almond oil*		*Citronella*		*p*-value R and SO	*p*-value SO and CO
		Mean	SD	Mean	SD	Mean	SD		
Good	20	54.75	16.66	57.40	17.92	68.90	20.97	0.516	0.004*
Bad	20	20.30	14.06	20.00	17.38	15.75	12.98	0.925	0.125
Active	20	49.40	19.03	50.30	30.43	50.60	15.03	0.901	0.962
Drowsy	20	38.20	24.42	40.85	22.66	34.30	22.64	0.669	0.343
Fresh	20	48.30	22.46	47.35	17.10	54.25	14.67	0.850	0.040*
Relax	20	52.20	23.80	50.85	22.38	71.15	16.24	0.804	0.002*
Stress	20	22.25	14.18	20.80	10.29	15.15	10.24	0.680	0.074
Uncomfortable	20	24.15	8.72	21.40	9.82	18.90	6.59	0.483	0.456
Romantic	20	34.40	3.16	37.50	6.90	43.20	9.32	0.381	0.228
Frustrated	20	20.30	8.71	17.15	8.81	11.75	2.55	0.263	0.115
Calm	20	46.45	6.97	52.85	4.95	65.85	17.50	0.229	0.048*
Disgust	20	12.31	15.20	18.85	10.10	14.55	9.55	0.089	0.248

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Citronella oil (CO)

EEG data

The mean and Standard Deviation (SD) of power value are presented in Table 20. There were noticeable changes of band power in alpha 1 waves that significantly increased during the citronella inhalation in all brains areas (p-value < 0.05), except for left anterior (p-value = 0.093). Conversely, band powers in alpha 2 waves were significantly increased in center (p-value = 0.029) and left posterior (p-value = 0.006). There were noticeable changes of power in beta waves that significantly increased during a citronella inhalation in all brains areas (p-value < 0.05) except right anterior (p-value = 0.093). No significant change was observed in the case of theta power (p-value > 0.05, data) shown in Table 20. In Figure 16, the topographic map shows after inhaling citronella compared with resting and sweet almond oil inhalation.

The alpha 1 wave power increased obviously in bilateral temporal and central areas whereas the power of alpha 2 wave increased mainly in posterior brain area. In addition, an increase of beta wave power was observed in anterior and posterior parts of the brain.

Table 20 Mean and SD power values in eye closed state, sweet almond oil and citronella oil inhalations.

Area	Theta Power (μV^2)						*p*-value R and SO	*p*-value SO and CO
	Eye close		*Sweet almond oil*		*Citronella*			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	3.18	0.46	2.99	0.41	3.31	0.64	0.137	0.316
right anterior	3.36	0.51	3.21	0.51	3.62	0.74	0.350	0.259
Center	4.76	0.62	4.63	0.62	5.12	0.90	0.500	0.244
left posterior	2.12	0.29	2.09	0.29	2.51	0.39	0.768	0.131
right posterior	2.22	0.35	2.20	0.35	2.48	0.51	0.811	0.246
Area	**Alpha 1 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and CO**
	Eye close		***Sweet almond oil***		***Citronella***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	8.31	2.76	7.71	2.42	8.93	2.89	0.140	0.093
right anterior	9.10	3.16	8.44	2.74	10.19	3.28	0.177	0.016*
Center	12.66	4.11	11.62	3.47	14.17	4.20	0.202	0.012*
left posterior	9.42	3.26	9.26	3.00	12.57	3.80	0.762	0.002*
right posterior	10.76	3.45	10.68	3.04	14.52	3.99	0.921	0.003*
Area	**Alpha 2 Power (μV^2)**						***p*-value R and SO**	***p*-value SO and CO**
	Eye close		***Sweet almond oil***		***Citronella***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	2.59	0.41	2.51	0.38	2.79	0.44	0.462	0.279
right anterior	2.68	0.42	2.59	0.39	2.97	0.47	0.446	0.142
Center	3.95	0.60	3.74	0.53	4.52	0.65	0.328	0.029*
left posterior	4.54	0.86	4.70	0.81	5.52	0.92	0.457	0.006*
right posterior	6.79	1.46	7.16	1.27	7.97	1.56	0.227	0.153
Area	**Beta Power (μV^2)**						***p*-value R and SO**	***p*-value SO and CO**
	Eye close		***Sweet almond oil***		***Citronella***			
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
left anterior	0.28	0.02	0.28	0.03	0.31	0.03	0.890	0.032*
right anterior	0.29	0.03	0.30	0.03	0.33	0.04	0.930	0.093
Center	0.37	0.03	0.36	0.04	0.43	0.05	0.878	0.003*
left posterior	0.32	0.03	0.32	0.04	0.39	0.04	0.944	0.000*
right posterior	0.35	0.04	0.36	0.04	0.42	0.05	0.860	0.000*

*Significant difference, *p*-value < 0.05 Rest (R), sweet almond oil (SO), Citronella oil (CO)

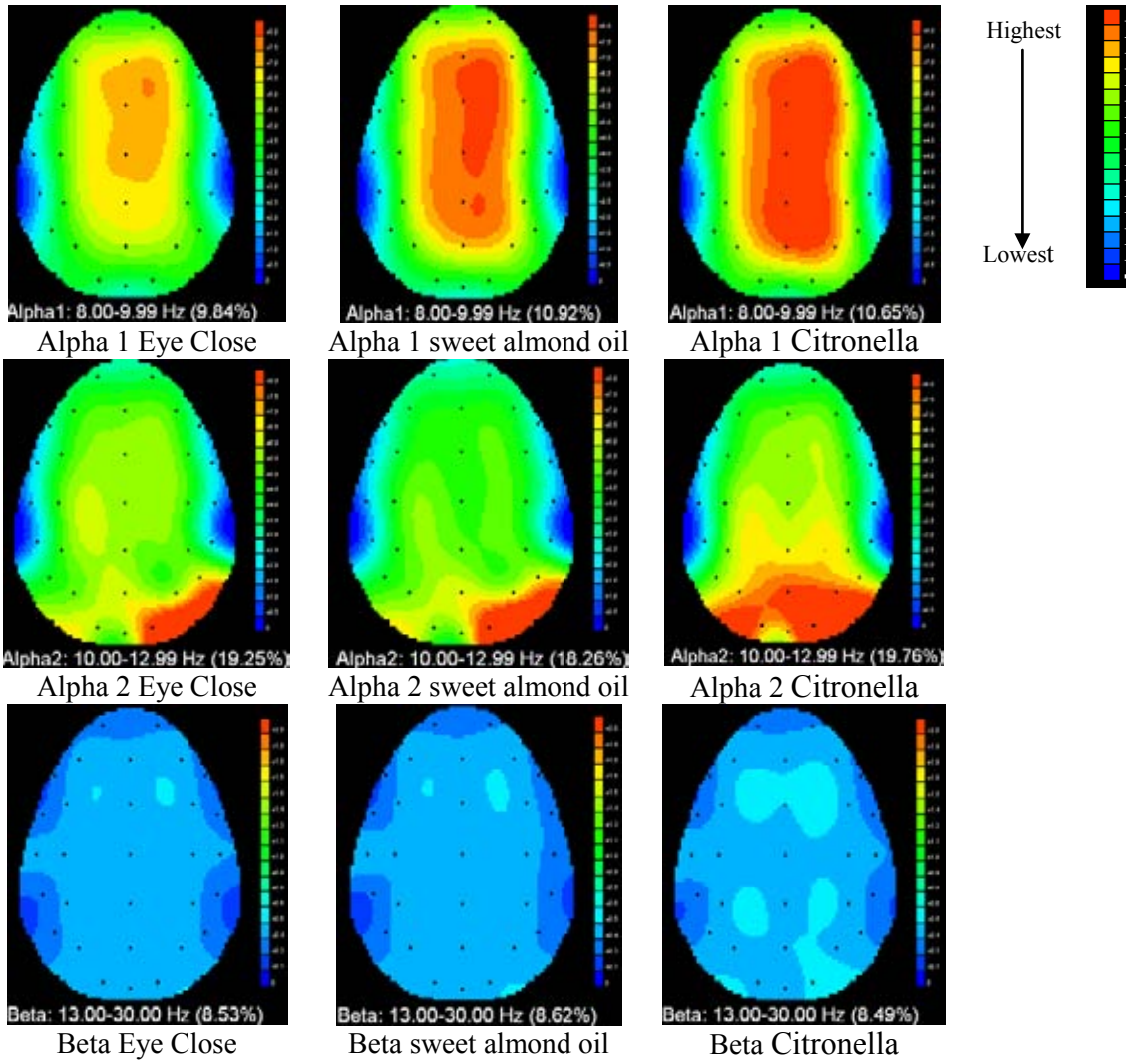


Figure16 The schematic diagram of the grand average of Alpha 1, Alpha 2, Beta topographical brain mapping in eye close, inhale sweet almond oil and inhale citronella.

The color bar represents the amount of energy in μV unit; maximum in red scale and minimum in blue scale. Topography range of both Alpha power 1 and 2, occurred during sweet almond oil inhalation comparing to citronella oil inhalation, increases from yellow to orange color. An increase of Beta power is observed by color changing from blue to light blue.

Correlation analysis

Table 21 revealed the correlation change between physiological parameters, including autonomic parameters, power brain wave and emotion by using Spearman rank order correlation coefficient for statistical analyses. Good emotion showed invert correlation with diastolic blood pressure (r = - 0.517, p = 0.019) ; fresh showed correlate with heart rate (r = 0.553, p = 0.004); romantic and calm showed negative correlation with respiratory rate (r = -0.577, p =0.004), (r = -0.575, p = 0.034) respectively. The correlation between brain power and mood state revealed that the change in calm emotion was correlated to right anterior alpha 1 (r = 0.560, p = 0.043); active motion revealed invert correlate in alpha 2 left posterior (r = -0.670, p = 0.033) but correlate with beta power (r = 0.590 , p = 0.030) . Otherwise, there was no significantly between physiological and emotional parameters.

Table 21 Correlation among changes of three parameters: autonomic parameters, brain wave power and emotional state after citronella oil inhalations.

Emotions	**ANS**	**Brain wave**		
		Alpha 1	**Alpha 2**	**Beta**
Good	Diastolic BP (r = -0.517*) p = 0.019			
Active			LP (r = -0.670*) p = 0.033	LA (r = 0.590*) p = 0.030
Fresh	HR (r = 0.553*) p = 0.004			
Romantic	RR (r = -0.577*) p = 0.033			
Calm	HR (r = -0.681*) p = 0.032 Respiratory rate (r = -0.575*) p = 0.034	RA (r = 0.560*) p = 0.043		

*Significant correlation, *p*-value < 0.05, r (Correlation coefficient), Diastolic BP (Diastolic blood pressure), HR (Heart rate), RR (Respiratory rate), LA (left anterior), RA (right anterior), LP (left posterior)

CHAPTER V

DISCUSSIONS

Effect of Lavender oil on physiological and emotions

In the present research, lavender oil was administered by inhalation to healthy subjects. Brain wave activity and ANS parameters (blood pressure, heart rate, respiratory rate and skin temperature) were recorded as indicators of the arousal level of nervous system. In addition, subjects had to rate their emotion terms of good, bad, active, drowsy, fresh, relaxed, stressed, uncomfortable, romantic, frustrated, calm, and disgusted in order to assess subjective behavioral arousal. Inhalation of lavender oil significantly decreased the level of ANS arousal, namely, decreases of blood pressure, heart rate and skin temperature. These changes of the ANS parameters represent the function of parasympathetic nervous system that counteracts the function of sympathetic nervous system. As for emotions, subjects felt better, fresher, more active, more relaxed, and less drowsy. This finding points towards a decrease of arousal as assessed through subjective self-evaluation. The results of this study support previous studies indicating lavender odor can influence relaxing. Previous studies using a footbath containing lavender oil also supports the positive effects on the parasympathetic neural activity of lavender oil [132]. To study the underlying mechanism of lavender oil on the nervous system, its main component, linalool, is used as a compound to study its effects compared with plain lavender oil. It is noteworthy that Heuberger and her colleagues found the reduction of blood pressure and skin temperature after applying linalool to the skin of participants [133]. In addition, linalool has a lot of isoforms in nature such as (*R*)-(−)-, (*S*)-(+)- and (*RS*)-(±)-forms. One study using R-(−)-linalool found similar effects from this compound on the autonomic nervous system parameters and also promoted calming and feelings of vigor [89]. According to the pharmacokinetic properties of linalool, Yamada was able to show the lipophilic properties of the linalool was suitable for transporting this compound across the blood-brain barrier. When reaching the brain, linalool can bind with the GABA (gamma aminobutyric acid) receptors similar to the benzodiazepines and caused relaxing and sedative effects [134]. In one study, they found linalool can potentiate the effects of GABA, the main inhibitor neurotransmitters of the human brain in the amygdala, the

subcortical brain area involved in the emotional response to the environment [135]. The effect of linalool on the amygdala may explain the emotion effects of lavender.

It is felt the effects of lavender inhalation on the brain wave activities are well demonstrated in this study. During inhalation with lavender, the power of theta (4-8 Hz) and alpha (8-13 Hz) activities are significantly increased in all brain regions. This result is consistent with the study of Diego which found after lavender inhalation that frontal alpha power was significantly increased [136]. Furthermore, a study conducted at the University of Occupational and Environmental Health, Kitakyushu Japan used changes of electroencephalogram (EEG) to measure the effects of aromas. This study found relaxing effects with increases of alpha wave activities after administering lavender, cineol sandalwood and alpha-pinene [137]. The EEG evidence of relaxation can be seen in various practices such as meditation. Meditation is a way of balancing the body and the mind as well as controlling the mind to experience feelings of peace and relaxation. The study among people meditating can demonstrate similar EEG changes with lavender inhalation which presented as an increase in theta and alpha activities in the brain during meditation [138]. The increase in theta and alpha activities can also be observed even during pre-meditation states in people who frequently practice meditation [139]. These results lend support that increases in theta and alpha wave activity causes a range of general relaxation effects and can be induced by a range of chemical and non-chemical techniques [140].

Effect of Rosemary oil on physiological and emotions

Presently researched, rosemary oil was inhaled by healthy subjects. Brain wave activity and ANS parameters (blood pressure, heart rate, respiratory rate and skin temperature) were recorded as indicators of the arousal level of nervous system. In addition, subjects had to rate their emotion in terms of good, bad, active, drowsy, fresh, relaxed, stressed, uncomfortable, romantic, frustrated, calm, and disgusted in order to assess subjective behavioural arousal. After rosemary oil inhalation, the result revealed stimulant sympathetic nervous system which induced the increase of heart rate, blood pressure, speeding of breathing rate and more muscle contraction, less blood circulation

which caused skin temperature significantly decreased. The results agreed with previous massage study using rosemary by increasing blood pressure and breathing rate [14]. Moreover, the results supported previous studies indicating that rosemary components contain abundant oxides (1, 8 cineol) and monoterpenes (α-pinene). Both components are the main action of stimulating the nervous system under sympathetic control. Previous studies demonstrated that these constituents remarkably has many biological activities such as Orhan *et al*. found 1, 8 cineole and α-pinene moderate inhibited of acetylcholenesterase which result in prolonged muscle contraction [141]. Heuberger's study showed 1, 8 cineole increased respiratory rate after administration substance [142]. As for emotional states, subjects felt better, more active, fresher, and less drowsy. This finding points towards an increasing of arousal as assessed through subjective self-evaluation. The result related to a medicinal benefit from which inhaled rosemary oil results in the removal boredom and gives fresh mental energy.An animal study found that after diffusion rosemary in the air encouraged the dogs spend more of their times alert (standing, moving) than lavender and chamomile [143]. Moss and colleague assess the olfactory impact of the essential oils of lavender and rosemary on cognitive performance and emotion in healthy volunteers [144]. They reported rosemary produced a significant enhancement of performance for overall quality of memory and secondary memory factors. With regard to emotion, comparisons of the change in ratings from baseline subjects significant more alert and fresh than the control group (no odor). Moreover after massage rosemary subject felt more vigorous and more cheerful [14]. The effects of rosemary inhalation on brain wave activities seemed well demonstrated in this study. During inhalation with rosemary, the power alpha 1 (8-10.99 Hz) and alpha 2 (11-12.99 Hz) activities are significantly decrease in all brain regions but the power beta (13-30 Hz) are considerably increase in frontal brain area. In this result concordance with pattern of EEG varies with the arousal level of CNS which increase in central activation is typically characterize by decrease in alpha and increase beta in contrast, sleep drowsy alpha increase and decrease beta [145]. This result consists of the study of Diego which found after rosemary inhalation that frontal alpha power was significantly increased [136]; however according to many areas attached with the electrode, the study also demonstrated the decrease of alpha 1 and alpha 2 value in posterior temporal region.

Furthermore, previous studies supported that cineol, the main constituent in rosemary, effected to brainwave change. For instance, Nakagawa *et al*. found methyl jasmonate and cineol, major components of jasmine oil and aromas effect on the increase of beta wave activity and inhibit the enhancement of alpha and theta waves which is a stimulating effect on the brain [106]. As a result, rosemary possibly contains 1, 8 cineol as its activities were similar to previous study.

Effect of jasmine oil on physiological and emotions

In the present research, jasmine oil was administered by inhalation to healthy subjects. Brain wave activity were recorded as indicators of the arousal level of central nervous system. In addition, subjects had to rate their emotional in terms of good, bad, active, drowsy, fresh, relaxed, stressed, uncomfortable, romantic, frustrated, calm, and disgusted in order to assess subjective behavioral arousal. These changes in the ANS parameters represent the function of sympathetic. These effects reflect the stimulating effects. The result demonstrated that subjects felt better, more active, fresher, more relax and more romantic after inhalation. Consequently, negative emotions such as drowsy, uncomfortable and disgust had been decreased in their feeling. The results also supported previous study referring jasmine odor induced stimulating effect. According to Tsuchiya *et al.* experiment, it found that jasmine and lemon oil responded to the sleeping time in mice by reducing the duration of sleep induced by barbiturate [103]. From Prachantasean study, the effect of essential oil on spatial learning in mice took a significant the shorter path length and time to find the hidden platform in the water when compare to the control group [146]. Our finding supported previous studies which indicating that after transdermal jasmine oil in forty healthy volunteers, it significantly increased blood pressure, breathing rate, more alert vigorous and less relax than those of the subjects in control group [15]. Accordingly the Holmes reported that jasmine oil cause the increase releasing of endorphin which is the substance that can promote many effect on nervous system [147]. During inhalation with jasmine oil, the power beta (13-30 Hz) considerably increased in frontal brain area. This result concordance with the pattern of EEG varies with the arousal level of CNS which increase in central activation is typically

characterize by increase beta [148]. This result was similar to the studies of Sugano and Nakagawa et al., found that jasmine odor is effect on increase of beta wave activity which is a stimulating effect on the brain. While methyl jasmonate, major component of jasmine oil, and cineol aromas inhibited the enhancement of alpha and theta waves which seemed to show a stimulating effect of the jasmine oil [102].

The result demonstrated that subjects felt better, more active, fresher, more relax and more romantic after inhalation. Consequently, negative emotions such as drowsy, uncomfortable and disgust had been decreased in their feeling. The results also supported previous study referring jasmine odor induced stimulating effect. To study underlying mechanism of main component of jasmine oil may also relevant that second messenger for some serotonin receptors is also cAMP and serotonin is certainly involved in the control of emotion within the central nervous system .The stimulant effect of inhale jasmine vapor is due to its absorptions and sequent pharmacological action within the brain or is merely due to the stimulation of odor receptor [149-150]. Our results demonstrated the stimulating/activating effect of jasmine oil and provide evidence for its use in aromatherapy for the relief of depression and uplifting emotion in humans. This study investigated the effects jasmine oil massage on menopausal symptoms in Korean climacteric women for 8 weeks. Kupperman's menopausal index was used to compare an experimental group of 25 climacteric women with a wait-listed control group of 27 climacteric women. The experimental group reported a significantly lower total menopausal index than wait-listed controls [151]. These findings suggest that aromatherapy massage may be an effective treatment of menopausal symptoms such as hot flushes, depression and pain in climacteric women.

Effect of Citronella oil on physiological and emotions

In the present study, citronella oil was administered by inhalation to healthy subjects. Brain wave activity and ANS parameters (blood pressure, heart rate, respiratory rate and skin temperature) were recorded as indicators of the arousal level of nervous system. In addition, subjects had to rate their emotion in terms of good, bad, active, drowsy, fresh, relaxed, stressed, uncomfortable, romantic, frustrated, calm, and disgusted

in order to assess subjective behavioral arousal. The results of this study support previous studies indicating citronella balancing effect. The observed effects of citronella are not precisely characterized by concept like stimulant or relaxation since inhalation of citronella oil significantly decreased the level of ANS arousal, decreases of blood pressure, heart rate and respiratory rate. As for emotions, subjects felt better, fresher, more relaxed, and more calm. The power alpha 1 (8-10.99 Hz), alpha 2 (11-12.99 Hz) and the power beta (13-30 Hz) activities were significantly increased. Since several reports had demonstrated about citronella on physiological effect, this finding was suggested obviously the influence on pharmacological effects. After analyzing by GCMS, there were 3 main components in citronella including terpine, geraniol, citronellol which are monoterpine alcohol in the same group of linalool and eugenol. For α–citronellal, it was classified into acyclic terpine aldehyde that was compared to a finding in discussion context herein. In general, monoterpene inhibits gamma-aminobuyric acid (GABA) tranaminase, thereby significantly increasing GABA level and decreasing glutamate level. Both of these alternations into the inhibitory and excitatory neurotransmitter systems are compatible relevant to the sedative effect [82]. Hamamota suggestd that the GABAergic transmission may be relevance for the mechanism of action of other monoterpenes such as α pinene,eugenol, citronellal, citronellal and hinokitol [152]. Azarmi *et al*. found that the vascular effect of geraniol by isolating rat aorta. Geraniol was able to reduce the contractile response to noradrenalin leading further rat aorta relaxing with low blood pressure and low heart rate [153]. In comparison with other volatile oils comprising similar components as those of citronella, such as rose oil *(Rosa Damascena* Mill) having main components of geraniol and citronellol, Khyaudeen indicated that 30 subjects who inhaled rose oil for 15 minutes had significantly blood pressure droping and relax emotion inducing. In EEG report, rose oil significantly decreased beta power but increased alpha power [154].

The change of beta wave after citronella inhalation was contradictory to an above report about changes of EEG relaxation. However, the significant increase of beta brainwave was related to high arousal level. Overall, the changes observed could be interpreted as refracting the harmonious status of arousal and relaxed, the so-called

"relaxed co ncentrate". C oincident r esearch on t his c hanges was obs erved, Hongratanaworakit tested on Ylang-Ylang o il e ffect characterized b y t he co ncept o f "harmonization" according t o p ost-oil i nhalation i nducing d ecreases o f blood pr essure and pul se r ate w hereas i ncrease of s ubjective emotions i ncluding a ttentive a nd a lert [117]. S imilarly, M orinushi studied on t he c ombination e ffect of P eppermint oi l a nd eucalyptus o il gave the s ame tendency findings [155]. Consequently, these results were possibly relevant in order to individually increase cognitive and mentally relaxing effect based on the evidences from the increase of alpha and beta brain powers.

Correlation analysis

In t able 22 s ummarize co rrelation an alysis b etween p hysiological p arameters, including a utonomic p arameters, po wer brain wave and e motion b y using Spearman rank- order correlation coefficient for statistical analyses.

Table 22 Significant correlation summary among change of three parameters: autonomic parameters, brain wave power and emotional state.

Emotion	ANS					Brain Wave		
	SB	DB	HR	RR	ST	Alpha1	Alpha2	Beta
Good		↓(CO)						
Active	↓(RO)		↑(JO)				↓ (CO)	↑ (CO)
Drowsy			↓(JO)		↑(LO)			↓(RO,JO)
Fresh			↑(CO)			↓ (RO)		↑ (LO)
Relax						↑ (RO,JO)	↑(RO,JO)	↓ (JO)
Stress							↓ (JO)	
Uncomfortable							↓ (JO)	
Calm		↓(LO)	↓(CO)	↓(CO)		↑(RO,JO, CO)	↑ (JO)	
Disgust						↓ (JO)	↓ (JO)	

The s pearman r ank co rrelation co efficient revealed t he r elationship b etween emotions w ith a utonomic ne rvous s ystem a nd pow er of br ain w ave i n e ach vol atile oi l

when compare to sweet almond oil (base oil) inhalation. Table symbols indicate as LO (lavender oil), RO (Rosemary oil), JO (Jasmine oil), CO (Citronella oil), (↑) significant positive correlation, (↓) significant negative correlation, SB (Systolic blood pressure), DB (Diastolic blood pressure), HR (Heart rate), RR (Respiratory rate) and ST (Skin temperature).

From the correlation in Table 22, it is obviously indicated that an efficacious mechanism of volatile oils to human can be classified into 2 ways including physiological and emotion effects. The mechanism of physiological effect directly activates both central nervous system and autonomic nervous system. On the other hand, emotion effect only activates through an olfactory nerve which is related between physiological and emotion effects.

For emotions and autonomic nervous system, an active feeling significantly increased systolic blood pressure in rosemary oil and heart rate in jasmine oil, furthermore, fresh emotions show a positive correlation with heart rate in citronella oil. Whereas good and calm feelings decreased a diastolic blood pressure in citronella oil and lavender oil respectively. Additionally, Clam feeling showed convert correlation with heart rate and respiratory rate in citronella oil. Moreover, drowsy feeling led a decrease on heart rate in jasmine oil. The relationship between body and emotion demonstrates clearly that active and fresh feelings were associated tightly to sympathetic system, leading to a secretion of norepinephrine from peripheral nerve. This further stimulates the increased action of SA node and increased heart rate, influencing on vascular constriction with high blood pressure. In contrast, good drowsy and calm feelings, which are parasympathetic system related, had influenced on acetylcholine secretion at vagus peripheral nerve, leading further to slower heart rate and generate a vasodilatation with low blood pressure release [53-54, 63-68, 83]

For a central nervous system, it is correlated with a frequency of brain waves. The emotion was shown that relate obviously to alpha wave (8-13Hz) and beta wave (13-30 Hz), corresponding to brain wave theory. The increase of alpha wave was observed whereas beta wave was decreased under relaxing state. On the other hand, stimulant or active condition activated beta wave increasingly while inactivated alpha wave [8, 58,

83]. Coincidently, volunteers, experienced to relax found an increase of alpha wave and a decrease of beta wave in rosemary and jasmine oil. In the other hand active and fresh emotions stimulated an increase of beta wave while increasing alpha wave. However, a change of power may occur in only one brain wave such as clam emotions. The increase of alpha wave, drowsy emotion showed a negative correlation with beta wave. In addition, uncomfortable and stress emotions inactivated the alpha wave in jasmine oil.

In conclusion, this research demonstrates the efficacy of popular volatile oils in Thailand on physiological effect and emotions. Aromatherapy is apparently a main target for the volatile oil application, various specifications of these products will be considered.

Future study

1. The future study effect aroma should focus on other volatile oils or measure the effect of each ingredient in various formula of volatile oils used in aromatherapy.
2. Brain wave determination using EEG revealed that the odor has influenced on brain waves. Then, the further study on ERP (Event related potential) should be performed to measure an ability of cognitive performance after volatile oil inhalation.
3. Research in field of aroma science not only do in laboratory, but also apply a volatile oil to , such as, athletes in order to stimulate sport performance, or to sleepless patient with a calm odor using suitable instruments for physiological monitoring.

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Appendices

Appendix A

Certified of Approval

AF 02-11

The Ethics Review Committee for Research Involving Human Research Subjects, Health Science Group, Chulalongkorn University
Institute Building 2, 4 Floor, Soi Chulalongkorn 62, Phyar hai Rd., Bangkok 10330, Thailand.
Tel: 0-2218-8147 Fax: 0-2218-8147 E-mail: eccu@chula.ac.th

COA No. 009/2011

Certificate of Approval

Study Title No.111.2/53 : **EFFECTS OF SELECTED VOLATILE OILS COMMONLY USED IN THAILAND ON PHYSIOLOGICAL ACTIVITIES AND MOOD STATES**

Principle Investigator : Mr. Winai Sayorwan

Place of Proposed Study/Institution : College of Public Health Sciences, Chulalongkorn University

The Ethics Review Committee for Research Involving Human Research Subjects, Health Science Group, Chulalongkorn University, Thailand, has approved constituted in accordance with the International Conference on Harmonization – Good Clinical Practice (ICH-GCP) and/or Code of Conduct in Animal Use of NRCT version 2000.

Signature: ..... Signature: .....

(Associate Professor Prida Tasanapradit, M.D.) (Assistant Professor Dr. Nuntaree Chaichanawongsaroj)

Chairman Secretary

Date of Approval : 24 January 2011 Approval Expire date : 23 January 2012

The approval documents including

1) Research proposal
2) Patient/Participant Information Sheet and Informed Consent Form
3) Researcher
4) Questionnaire

Protocol No. 111.2/53
Date of Approval 24 JAN 2011
Approval Expire Date 23 JAN 2012

The approved investigator must comply with the following conditions:

1. The research/project activities must end on the approval expired date of the Ethical Review Committee for Research Involving Human Research Subjects, Health Science Group, Chulalongkorn University (ECCU). In case the research/project is unable to complete within that date, the project extension can be applied one month prior to the ECCU approval expired date.
2. Strictly conduct the research/project activities as written in the proposal.
3. Using only the documents that bearing the ECCU's seal of approval with the subjects/volunteers (including subject information sheet, consent form, invitation letter for project/research participation (if available); and return the first subject's copy of the above documents to the ECCU.
4. Report to the ECCU for any serious adverse events within 5 working days
5. Report to the ECCU for any change of the research/project activities prior to conduct the activities.
6. Final report (AF 03-11) and abstract is required for a one year (or less) research/project and report within 30 days after the completion of the research/project. For thesis, abstract is required and report within 30 days after the completion of the research/project.
7. Annual progress report is needed for a two- year (or more) research/project and submit the progress report before the expire date of certificate. After the completion of the research/project processes as No. 6.

Appendix B

หนังสือแสดงความยินยอมเข้าร่วมคัดเลือกเป็นอาสาสมัครในการวิจัย (ภาษาไทย)

เลขที่ ประชากรตัวอย่างหรือผู้มีส่วนร่วมในการวิจัย…..……

ข้าพเจ้า ซึ่งได้ลงนามท้ายหนังสือนี้ ขอแสดงความยินยอมเข้าร่วมโครงการวิจัย

ชื่อโครงการวิจัย: ผลของน้ำมันระเหยบางชนิดที่ใช้มากในประเทศไทย ต่อสรีรวิทยา และอารมณ์ความรู้สึก

ชื่อผู้วิจัย นาย วินัย สยอวรรณ นิสิตปริญญาเอก วิทยาลัยวิทยาศาสตร์สาธารณสุข จุฬาลงกรณ์มหาวิทยาลัย
ที่อยู่ที่ติดต่อที่ทำงาน: วิทยาลัยวิทยาศาสตร์สาธารณสุข จุฬาลงกรณ์มหาวิทยาลัย
ชั้น 10 อาคารสถาบัน 3 ซ. จุฬาฯ 62 ถ.พญาไท แขวงวังใหม่ เขตปทุมวัน กทม. 10330
ที่อยู่ที่ติดต่อที่บ้าน: 626/2 ซอยริมคลองชักพระ แขวงคลองชักพระ เขตตลิ่งชัน กรุงเทพ 10170
โทรศัพท์ (ที่ทำงาน) 0-2218-8152-3 โทรศัพท์ที่บ้าน 086-5729771
โทรศัพท์มือถือ 086-5729771 E-mail address: winsayyes @hotmail.com

ข้าพเจ้า **ได้รับทราบ** รายละเอียดเกี่ยวกับที่มาและวัตถุประสงค์ในการทำวิจัย รายละเอียดขั้นตอนต่างๆ ที่จะต้องปฏิบัติหรือได้รับการปฏิบัติ ความเสี่ยง/อันตราย และประโยชน์ซึ่งจะเกิดขึ้นจากการวิจัยเรื่องนี้ โดยได้อ่านรายละเอียดในเอกสารชี้แจงผู้เข้าร่วมการวิจัยโดยตลอด และ**ได้รับคำอธิบาย**จากผู้วิจัย **จนเข้าใจเป็นอย่างดี**แล้ว

ข้าพเจ้ายินดีเข้าร่วมเพื่อการคัดเลือกเป็นอาสาสมัครในการวิจัยครั้งนี้ ภายใต้เงื่อนไขที่ระบุไว้ในเอกสารข้อมูลสำหรับกลุ่มประชากรหรือผู้มีส่วนร่วมในการวิจัย และยินยอมรับการคัดเลือกโดยต้องมีการประเมินสุขภาพโดยแบบสอบถาม ประเมินความสามารถในการรับกลิ่น ประเมินข้างของมือที่ถนัดในการใช้งาน ประเมินความชอบกลิ่น

ข้าพเจ้าจึง**สมัครใจ**เข้าร่วมในโครงการวิจัยนี้ ตามที่ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย โดยข้าพเจ้ามีสิทธิ**ถอนตัว**ในการเป็นอาสาสมัคร เมื่อใดก็ได้ตามความประสงค์ **โดยไม่ต้องแจ้งเหตุผล** ซึ่งการถอนตัวออกจากการเป็นอาสาสมัครนั้น จะไม่มีผลกระทบในทางใดๆ ต่อข้าพเจ้าทั้งสิ้น

ข้าพเจ้าได้รับคำรับรองว่า ผู้วิจัยจะปฏิบัติต่อข้าพเจ้าตามข้อมูลที่ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย และข้อมูลใดๆ ที่เกี่ยวข้องกับข้าพเจ้า ผู้วิจัยจะ**เก็บรักษาเป็นความลับ** โดยจะนำเสนอข้อมูลการวิจัยเป็นภาพรวมเท่านั้น ไม่มีข้อมูลใดในการรายงานที่จะนำไปสู่การระบุตัวข้าพเจ้า

หากข้าพเจ้าไม่ได้รับการปฏิบัติตรงตามที่ได้ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย ข้าพเจ้าสามารถร้องเรียนได้ที่คณะกรรมการพิจารณาจริยธรรมการวิจัยในคน กลุ่มสหสถาบัน ชุดที่ 1 จุฬาลงกรณ์มหาวิทยาลัย ชั้น 4 อาคารสถาบัน 2 ซอยจุฬาลงกรณ์ 62 ถนนพญาไท เขตปทุมวัน กรุงเทพฯ 10330
โทรศัพท์ 0-2218-8147 โทรสาร 0-2218-8147 **E-mail: eccu@chula.ac.th**

ข้าพเจ้าได้ลงลายมือชื่อไว้เป็นสำคัญต่อหน้าพยาน ทั้งนี้ข้าพเจ้าได้รับสำเนาเอกสารชี้แจงผู้เข้าร่วมการวิจัย และสำเนาหนังสือแสดงความยินยอมไว้แล้ว

ลงชื่อ.....	ลงชื่อ.....
(.....)	(.....)
ผู้วิจัยหลัก	ผู้มีส่วนร่วมในการวิจัย

Consent Form for selected subjects (English Version)

Date.....Month.....A.D.....

No. a population sample or a participant…..……

I, who have signed at the end of this document, consent to participate the research project.

Research topic: Effects of selected volatile oils commonly used in Thailand on physiological activities and emotions

Researcher: Mr. Winai Sayorwan, Ph.D. student of College of Public Health Science Chulalongkorn University

Address: 10th floor Institute 3Building Soi Chulalongkorn62 payathai road, Patumwan, Bangkok 10330

Home address: 626/2 Soi Rimklongchak pra ,Talingchan Bangkok 10170

Tel. (Office) 0-2218-8152-3 (Home) 086-5729771

Mobile phone: 086-5729771 E-mail address: winsayyes @gmail.com

I have known the detail of source and objective of the research, protocol detail to perform or to be performed, risk/hazard and advantage happened from this research by reading document information in detailed information for participants thoroughly. I am also described from a researcher until get well understanding.

I am pleased to participate in order to this selection as a volunteer under the condition designated in information for population group or research participants . And, I consent to receive the selection by health assessment using questionnaire that assess ability of odor smelling, assess right/left-handed side to work and assess odor favorite.

I am willing to participate to this research according to that designated in detail formation for participant document. I am able to withdraw from a volunteer whenever as wish by no informing. This withdrawal from a volunteer will not affect any ways to me absolutely.

I am guaranteed that researcher will act to me according to information designated in detailed information for participants. Moreover, researcher will keep my information or any information related to me in private and a secret. There will be no report subjected to my verification.

If I have not been performed in accordance with which designated in information for participants, I can complain to The Ethic review Committee for Research Involving Human Research Subjects ,Health Science Group, Chulalongkorn University (4th floor Institute 2Building Soi Chulalongkorn62 payathai road, Patumwan, Bangkok 10330) Tel. 0-2218-8147 Fax. 0-2218-8147 E-mail: eccu@chula.ac.th

I have signed at most in front of a witness, so as to that I have been already received a copy of information for participants and a copy of the consent form .

..... Signature	..... Signature
(.....)	(.....)
Main researcher	Participant

Appendix C

หนังสือแสดงความยินยอมเข้าร่วมการวิจัย (ภาษาไทย)

วันที่.....เดือน.....พ.ศ. .....

เลขที่ ประชากรตัวอย่างหรือผู้มีส่วนร่วมในการวิจัย.….....……

ข้าพเจ้า ซึ่งได้ลงนามท้ายหนังสือนี้ ขอแสดงความยินยอมเข้าร่วมโครงการวิจัย

ชื่อโครงการวิจัย: ผลของน้ำมันระเหยบางชนิดที่ใช้มากในประเทศไทย ต่อสรีรวิทยา และอารมณ์ความรู้สึก

ชื่อผู้วิจัย นาย วินัย สยอวรรณ นิสิตปริญญาเอก วิทยาลัยวิทยาศาสตร์สาธารณสุข จุฬาลงกรณ์มหาวิทยาลัย

ที่อยู่ที่ติดต่อที่ทำงาน: วิทยาลัยวิทยาศาสตร์สาธารณสุข จุฬาลงกรณ์มหาวิทยาลัย

ชั้น 10 อาคารสถาบัน 3 ซ. จุฬาฯ 62 ถ.พญาไท แขวงวังใหม่ เขตปทุมวัน กทม. 10330

ที่อยู่ที่ติดต่อที่บ้าน: 626/2 ซอยริมคลองชักพระ แขวงคลองชักพระ เขตตลิ่งชัน กรุงเทพ 10170

โทรศัพท์ (ที่ทำงาน) 0-2218-8152-3 โทรศัพท์ที่บ้าน 086-5729771

โทรศัพท์มือถือ 086-5729771 E-mail address: winsayyes @hotmail.com

ข้าพเจ้า ได้รับทราบรายละเอียดเกี่ยวกับที่มาและวัตถุประสงค์ในการทำวิจัย รายละเอียดขั้นตอนต่างๆ ที่จะต้องปฏิบัติหรือได้รับการปฏิบัติ ความเสี่ยง/อันตราย และประโยชน์ซึ่งจะเกิดขึ้นจากการวิจัยเรื่องนี้ โดยได้อ่านรายละเอียดในเอกสารชี้แจงผู้เข้าร่วมการวิจัยโดยตลอด และได้รับคำอธิบายจากผู้วิจัย จนเข้าใจเป็นอย่างดีแล้ว

ข้าพเจ้าจึงสมัครใจเข้าร่วมในโครงการวิจัยนี้ ตามที่ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย โดยข้าพเจ้ามีสิทธิถอนตัวออกจากการวิจัยเมื่อใดก็ได้ตามความประสงค์ โดยไม่ต้องแจ้งเหตุผล ซึ่งการถอนตัวออกจากการวิจัยนั้นจะไม่มีผลกระทบในทางใดๆ ต่อข้าพเจ้าทั้งสิ้น

ข้าพเจ้าได้รับคำรับรองว่า ผู้วิจัยจะปฏิบัติต่อข้าพเจ้าตามข้อมูลที่ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย และข้อมูลใดๆ ที่เกี่ยวข้องกับข้าพเจ้า ผู้วิจัยจะเก็บรักษาเป็นความลับ โดยจะนำเสนอข้อมูลการวิจัยเป็นภาพรวมเท่านั้น ไม่มีข้อมูลใดในการรายงานที่จะนำไปสู่การระบุตัวข้าพเจ้า

หากข้าพเจ้าไม่ได้รับการปฏิบัติตรงตามที่ได้ระบุไว้ในเอกสารชี้แจงผู้เข้าร่วมการวิจัย ข้าพเจ้าสามารถร้องเรียนได้ที่คณะกรรมการพิจารณาจริยธรรมการวิจัยในคน กลุ่มสหสถาบัน ชุดที่ 1 จุฬาลงกรณ์มหาวิทยาลัย ชั้น 4 อาคารสถาบัน 2 ซอยจุฬาลงกรณ์ 62 ถนนพญาไท เขตปทุมวัน กรุงเทพฯ 10330

โทรศัพท์ 0-2218-8147 โทรสาร 0-2218-8147 E-mail: eccu@chula.ac.th

ข้าพเจ้าได้ลงลายมือชื่อไว้เป็นสำคัญต่อหน้าพยาน ทั้งนี้ข้าพเจ้าได้รับสำเนาเอกสารชี้แจงผู้เข้าร่วมการวิจัย และสำเนาหนังสือแสดงความยินยอมไว้แล้ว

ลงชื่อ.....	ลงชื่อ.....
(.....)	(.....)
ผู้วิจัยหลัก	ผู้มีส่วนร่วมในการวิจัย

Consent Form (English Version)

Date.....Month.....A.D.....

No. a population sample or a participant…..……

I, who have signed at the end of this document, consent to participate the research project. Research topic: Effects of selected volatile oils commonly used in Thailand on physiological activities and emotions

Researcher: Mr. Winai Sayorwan, a Ph.D. student of College of Public Health Science Chulalongkorn University

Address: 10th floor Institute 3Building Soi Chulalongkorn62 payathai road, Patumwan, Bangkok 10330

Home address: 626/2 Soi Rimklongchak pra ,Talingchan Bangkok 10170

Tel. (Office) 0-2218-8152-3 (Home) 086-5729771

Mobile phone: 086-5729771 E-mail address: winsayyes @gmail.com

I have known the detail of source and objective of the research, protocol detail to perform or to be performed, risk/hazard and advantage happened from this research by reading document information in detailed information for participants thoroughly. I am also described from a researcher until get well understanding.

I am willing to participate to this research according to that designated in detail formation for participant document. I am able to withdraw from a volunteer whenever as wish by no informing. This withdrawal from a volunteer will not affect any ways to me absolutely.

I am guaranteed that researcher will act to me according to information designated in detailed information for participants. Moreover, researcher will keep my information or any information related to me in private and a secret. There will be no report subjected to my verification.

If I have not been performed in accordance with which designated in information for participants, I can complain to The Ethic review Committee for Research Involving Human Research Subjects ,Health Science Group, Chulalongkorn University

(4th floor Institute 2Building Soi Chulalongkorn62 payathai road, Patumwan, Bangkok 10330

Tel. 0-2218-8147 Fax. 0-2218-8147 E-mail: eccu@chula.ac.th

I have signed at most in front of a witness, so as to that I have been already received a copy of information for participants and a copy of the consent form .

..... Signature ..... Signature

(.....) (.....)

Main researcher Participant

Appendix D

แบบสอบถามข้อมูลสุขภาพ (ภาษาไทย)

โปรดตอบแบบสอบถามต่อไปนี้ ตามข้อมูลที่เป็นจริง ตรงกับตัวท่านมากที่สุด

1 ข้อมูลส่วนบุคคล

ชื่อสกุล ..... เพศ.....

อายุ ..... ปี น้ำหนัก..... ส่วนสูง..... ถนัดมือข้างไหน.....

เบอร์โทรศัพท์ที่สามารถติดต่อได้สะดวก .....

2 ข้อมูลด้านสุขภาพ

1. ท่านมีโรคประจำตัวดังต่อไปนี้ หรือไม่

- โรคทางระบบประสาท

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคลมชัก

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคติดเชื้อต่างๆ

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคติดเชื้อของระบบทางเดินหายใจ/ที่เกี่ยวข้องกับทางเดินหายใจ

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคหอบหืด

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคภูมิแพ้

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคไซนัส

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

- โรคความดันโลหิต

△ เป็น △ไม่เป็น ความดันโลหิตที่วัดได้ .....

- ท่านมีโรคประจำตัวอื่น คือ ..... และ/หรือ เคยเข้ารับการผ่าตัด .....

- ท่านจำเป็นต้องใช้ยารักษาโรคประจำตัว คือ

ชนิด ขนาด ปริมาณ

....

....

2. ท่านคิดว่าสุขภาพร่างกายของท่านเป็นอย่างไร

△ เจ็บป่วย △ ปกติตามเคย △แข็งแรงดี △ แข็งแรงดีมาก

3. ท่านเคยแพ้สิ่งต่อไปนี้ หรือไม่

△ สารเคมี..... △อาหาร..... △น้ำหอม △เกสรดอกไม้ △อื่นๆ

4. ท่านเคยประสบอุบัติเหตุร้ายแรง หรือไม่ △ เคยที่อวัยวะ..... เมื่อ..... △ไม่เคย

5. เวลานอนตามปกติ .....ชั่วโมง

6. ท่านมีปัญหาเรื่องนอนหลับในช่วง 1 เดือนที่ผ่านมา หรือไม่

△ เป็น △ไม่เป็น △ไม่ทราบ / ไม่แน่ใจ

7. ท่านมีปัญหาการได้ยิน หรือไม่ △ มี △ไม่มี

8. ท่านมีปัญหาในการดมกลิ่น หรือไม่ △ มี △ไม่มี

9. ท่านได้รับการฝังเครื่องกระตุ้นหัวใจ △ มี △ไม่มี

10. ท่านคิดว่าสุขภาพจิตของท่านเป็นอย่างไร

△ เจ็บป่วย △ ไม่ดี △ ดี

11. ท่านสูบบุหรี่หรือไม่

△ ไม่เคยเลย △ สูบ △ เคยสูบแต่หยุดสูบแล้ว

12. ท่านดื่มสุรา เครื่องดื่มที่มีแอลกอฮอล์หรือไม่

△ ไม่เคยเลย △ บ่อยครั้ง △ บางครั้ง

- น้ำอัดลม △ ใช่ ประมาณวันละ..... △ไม่ใช่ △ บางครั้ง - -ชา

กาแฟ △ ใช่ ประมาณวันละ..... △ไม่ใช่ △ บางครั้ง ….

ขอบพระคุณครับ

Health Status (English Version)

Please answer this questionnaire with honesty

1 Personal information

Age .....Wight..... Kg Height .....Cm Telephone Number.....

2 Health Information

1 Do you have these following illness or not?

-Neurological diseases.

△ Yes △No △ Not that I know/unsure…….

-Epilepsy

△ Yes △No △ Not that I

- Infection

△ Yes △No △ Not that I

- Asthma

△ Yes △No △ Not that I know/unsure

- Allergy

△ Yes △No △ Not that I know/unsure

-Sinus

△ Yes △No △ Not that I know/unsure

-High/ Low Blood Pressure

△ Yes △No △ Not that I know/unsure

Do you have other congenital disease is ..... And / or having to get surgery.

Are you on any regular medication?.....

2 How good is your health?

△ Sick △ Normal △ Healthy △ Very healthy

3. Have you ever allergic to these follows?

△ Chemical △ Food..... △ Perfume..... △ Pollens.....

4. Have you ever experienced any critical accident?

△ Yes, internally..... If yes, when?..... △ never

5. How long do you normally sleep a night?.....Hours

6. Do you have any sleeping problem during this past month?

△ Yes △No

7 Do you have any hearing problem?

△ Yes △No

8 Do you have any smelling disorder?

△ Yes △No

9 Have you been installed any pacemaker?

△ Yes △No

10 How is your mental health?

△ Sick △ Not well △ ok △ Good △ Very good

11. Have you ever smoked cigarette?

△ Never △ Yes △ Yes, but not anymore

12. Do you drink alcohol?

△ No △ Consistently △ Consistently but quit already

13. Do you drink these follows regularly?

- Pop soda

△ Yes, what's the quantity per day?..... △No △ Sometimes, How often?.....

- Tea, Coffee

△ Yes, what's the quantity per day?..... △No △ Sometimes

-Tonic beverage

△ Yes, what's the quantity per day?..... △No △ Sometimes

Appendix E

แบบทดสอบถนัดมือขวา (ภาษาไทย)

ให้ท่านอ่านกิจกรรมในแต่ละข้อ แล้วให้คะแนนตามความเป็นจริงมากที่สุด **<u>โปรดตอบทุกข้อ</u>** ตามมือข้างที่ถนัด หากไม่เคยมีประสบการณ์ในกิจกรรมนั้นๆให้เว้นช่องว่างไว้

<u>วิธีการให้คะแนน</u>

\+ ในช่องมือข้างที่ถนัดขณะทำกิจกรรมนั้น ซึ่งมืออีกข้างพอที่จะทำได้บ้าง

++ ในช่องมือที่ถนัดข้างเดียวโดยที่มืออีกข้างที่ไม่สามารถทำกิจกรรมนั้นได้เลย

+/+ ในทั้ง 2 ช่องถ้าสามารถทำกิจกรรมในแต่ละข้อนั้นได้ดี ทั้ง 2 มือเท่าๆกัน

กิจกรรม	ข้างขวา	ข้างซ้าย
1.เขียนหนังสือ		
2. วาดรูป		
3. โยน หรือ ปาของ		
4. ใช้กรรไกร		
5. ถือแปรงสีฟัน		
6. ถือมีดหั่นของ		
7. ถือช้อน		
8. กวาดพื้น		
9. ถือก้านไม้ขีดไฟ		
10. มือข้างที่ถือฝา ขณะเปิดฝากล่องหรือขวด		
คะแนนรวม		

..... ผู้ประเมิน

การคิดคะแนน

<u>ผลรวมของช่องข้างขวา –ช่องข้างซ้าย</u> ×100

ผลรวมทั้งหมด

เกณฑ์

ได้คะแนน ต่ำกว่า -40 แสดงว่าถนัดมือซ้าย

ได้คะแนน ระหว่าง -40- +40 แสดงว่าถนัดทั้งสองข้าง

ได้คะแนน มากกว่า +40 แสดงว่าถนัดข้างขวา

Edinburgh Handedness Inventory Test (English version)

Please indicate your preferences in the use of hands in the following activities by putting +in the appropriate column. Where the preference is so strong that you would never try to use the other hand, unless absolutely forced to, put ++ checks. If in any case you are really indifferent, put + in both columns.

Some of the activities listed below require the use of both hands. In these cases, the part of the task, or object, for which hand preference is wanted is indicated in parentheses.

Please try and answer all of the questions, and only leave a blank if you have no experience at all with the object or task.

Activity	Right Side	Left Side
1 Writing		
2. Drawing		
3 Throwing		
4. Scissors		
5. Toothbrush		
6. Knife (without fork)		
7. Spoon		
8. Broom (upper hand)		
9. Striking Match (match)		
10. Opening box (lid)		

Scoring:

Add up the number of checks in the "Left" and "Right" columns and enter in the "TOTAL" row for each column. Add the left total and the right total and enter in the "Cumulative TOTAL" cell. Subtract the left total from the right total and enter in the "Difference" cell. Divide the "Difference" cell by the "Cumulative TOTAL" cell (round to 2 digits if necessary) and multiply by 100; enter the result in the "Result" cell.

Below -40 = left-handed Between -40 and +40 = ambidextrous
above +40 = right-handed

Appendix F

Score sheet for odor test (Butanol Threshold)

Step	**Concentration**	**1**	**2**	**3**	**4**	**5**
11	(2.25×10^{-5})	B	W	B	B	W
10	(6.77×10^{-5})	B	B	w	w	B
9 Start	(2.03×10^{-4})	W	B	w	B	B
8	(6.09×10^{-4})	W	B	w	B	B
7	(1.82×10^{-3})	W	W	B	B	B
6	(5.48×10^{-3})	B	W	B	B	B
5	(0.0164%)	B	B	B	B	W
4	(0.049%)	W	B	B	B	W
3	(0.148%)	W	B	B	B	B
2	(0.44%)	W	B	B	B	B
1	(1.33%)	B	W	B	B	W
0	(4%)	B	W	B	B	W
Score						

B = Smell Buthanol W= Smell Water

Key : ✓ Correct × Incorrect

Appendix G

Odor Familiarity

Have you ever had these symptoms after inhalation? (Answer more than one item)

☐ Headaches / Dizziness ... ... ……☐ Rash……

☐ Runny nose ... ...☐ Allergy ……

☐ Breathing difficulty…….....☐ None had any symptoms……

☐ Nausea / Vomiting ... ...

How do you feel the smell of the following?

Score / Odor	Very much like 5	Like 4	Moderately 3	Don' t Like 2	Hate 1
1					
2					
3					
4					
5					

Appendix H

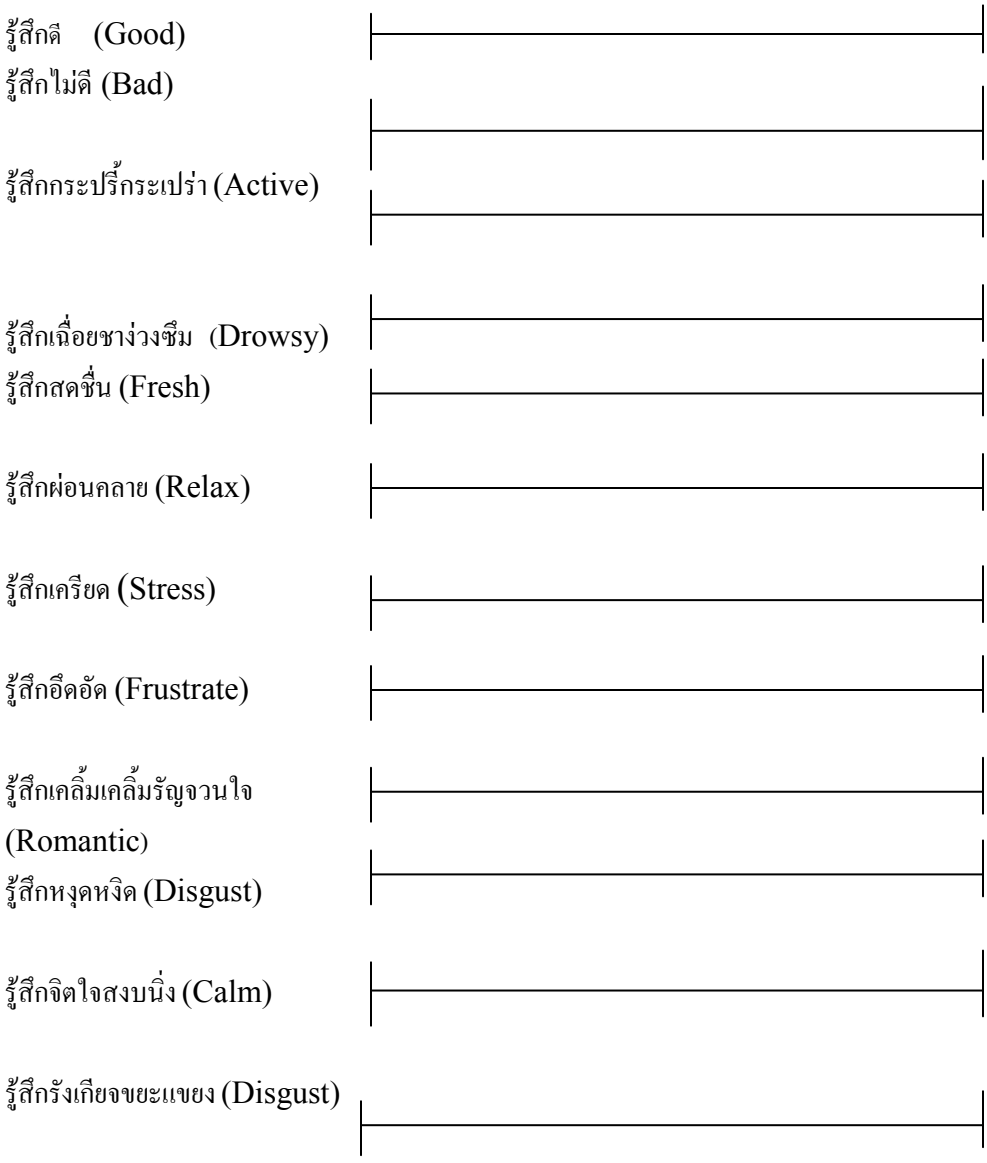
Case record Autonomic Nervous System

Activity		Times	Blood Pressure		Heart rate	Temp	RR	Note
			Systolic	Diastolic				
	1							
	2							
	3							
	4							
	5							
	6							
	7							
	8							
	9							
	10							
	1							
	2							
	3							
	4							
	5							
	6							
	7							
	8							
	9							
	10							
	11							
	12							
	13							
	14							
	15							

Appendix I

Emotional Record

ในนาทีนี้ท่านมีความรู้สึกตามหัวข้อต่อไปนี้อย่างไร ให้ท่านทำเครื่องหมาย │ ลงบนเส้น จากน้อยไปหามาก



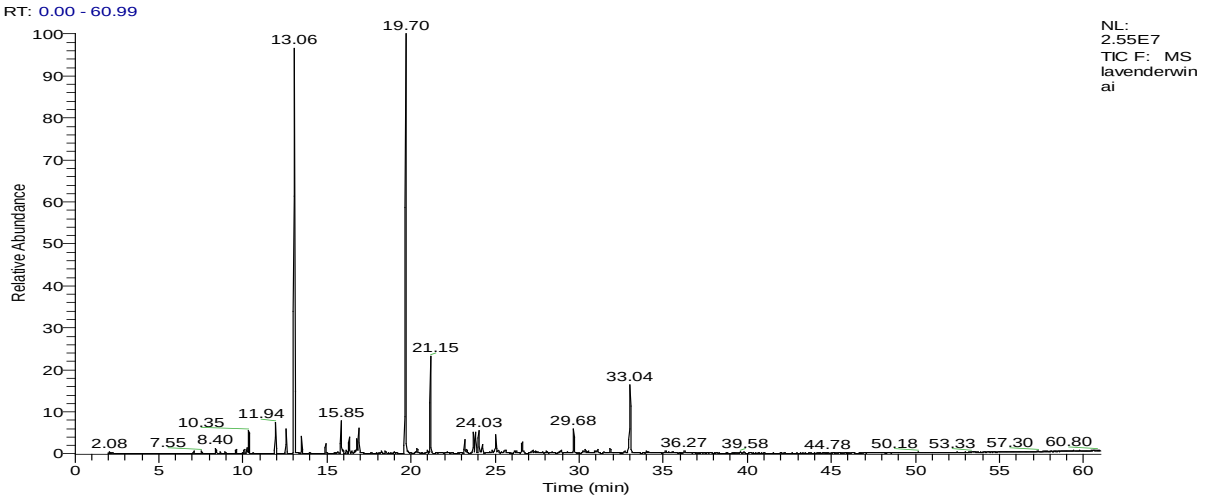
ท่านมีอาการข้างเคียง หลังดมกลิ่นหรือไม่ ระบุ .....

Have you ever had these symptoms after inhalation.....

Appendix J

GC Chromatogram

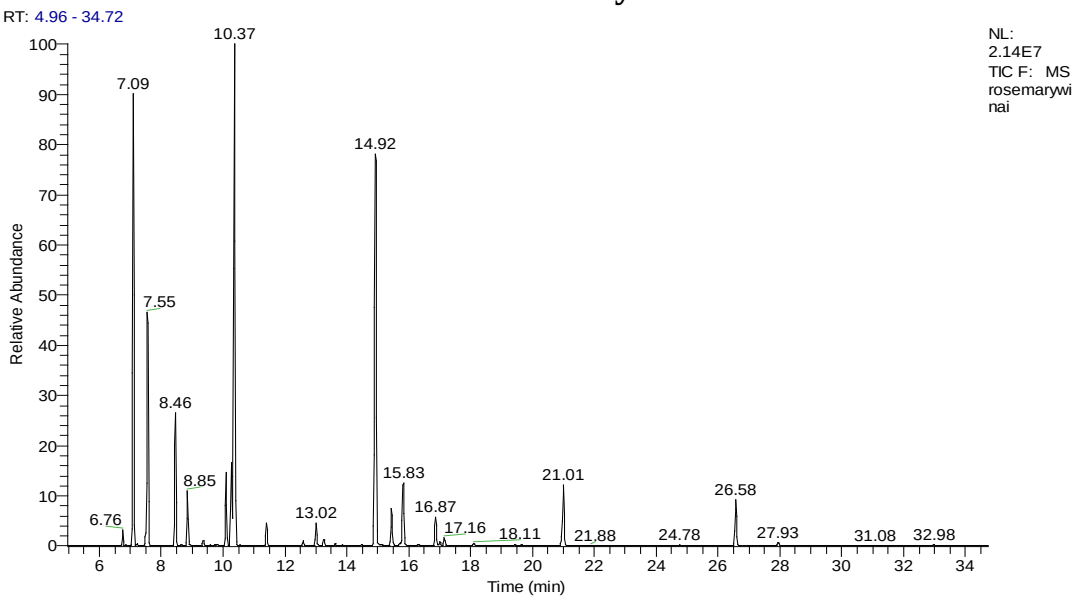
Lavender oil



RT	**Name Compound**	**Kovat's Index**	Area %
10.35	Eucalyptol	1187.91	1.46
11.94	oxiranemethanol	1373.54	1.86
12.56	4-methyl-3-pentenyl]oxiranemethanol	1227.00	1.70
13.06	Alpha Linalool	4963.75	31.91
13.49	Octen-1-ol	1018.19	1.09
15.85	Borneol	1409.47	2.61
16.89	Alpha -Terpieol	1230.53	1.91
19.70	Linalyl acetate	5047.98	32.46
21.15	Lavandulyl acetate	2427.74	6.82
23.72	Limonene oxide	1126.16	1.42
23.84	2-Octen-1-ol, 3,7-dimethyl-, isobutyrate	1119.21	1.48
24.03	3,7-Octadiene-2,6-diol, 2,6-dimethyl	1163.07	1.72
25.04	Neryl acetate	1061.39	1.77
33.04	Caryophyllene oxide	2031.74	6.20

Figure 17 GC chromatogram of lavender oil

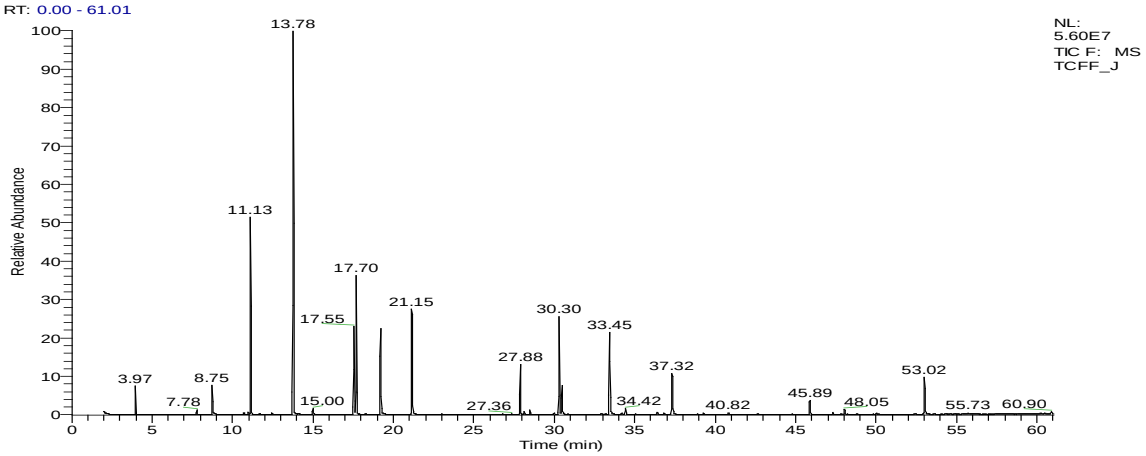
Rosemary oil



RT	**Name Compound**	**Kovat's Index**	Area %
7.09	Alpha -pinene	4388.95	19.41
7.55	Camphene	3151.94	9.81
8.46	Beta-myrcene	2380.79	4.88
8.85	Beta -Pinene	1525.42	1.96
10.10	o-Cymene	1771.50	2.00
10.26	Limonene	1876.00	3.01
10.37	1,8 Cineole	4621.81	20.08
11.40	1,4-Cyclohexadiene	977.20	0.92
13.02	Alpha-Linalool	988.16	1.08
14.92	Camphor	4085.72	21.25
15.44	Isoborneol	1250.52	1.79
15.83	Borneol	1635.46	3.29
16.87	Alpha -Terpineol	1102.69	1.58
21.01	Isobornyl acetate	1604.94	2.00
26.58	Caryophyllene	1403.59	2.34
47.04	Diisooctyl phthalate	909.48	4.26

Figure 18 GC chromatogram of rosemary oil

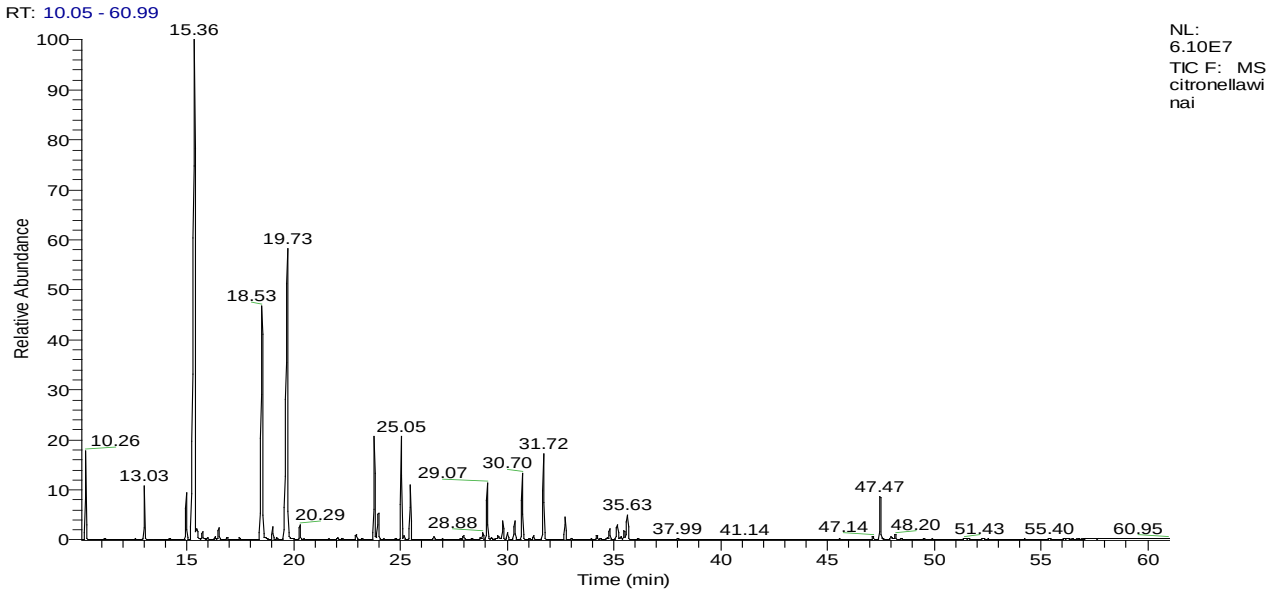
Jasmine Oil



RT	Name Compound	Kovat's Index	Area %
3.97	Hexenol	867	1.11
8.75	Benzyl alcohol	1032	1.92
11.13	Beta-Linalool	1098	11.22
13.78	Benzyl acetate	1163	26.09
17.55	Linalyl acetate	1257	5.65
17.70	Benzyl propionate	1257	9.65
19.21	Indole	1288	6.68
21.15	Methyl anthranilate	1337	8.54
27.88	Alpha-Farnesene	1508	3.42
30.30	cis-3-Hexenyl Benzoate	1570	7.09
30.47	Gamma-Cadinol	1645	2.41
33.45	cis-Methyl dihydrojasmonate	1654	6.47
34.42	Trans-Methyl dihydrojasmonate	1680	0.46
37.32	Benzyl Benzoate	1762	3.59
45.89	trans, trans-Farnesyl acetate	1843	1.14
48.05	Methyl linolenate	2092	0.44
53.02	9-Tricosene	2315	3.09

Figure 19 GC chromatogram of jasmine oil

Citronella oil



RT	Name Compound	Kovat's Index	Area %
10.26	D-Limonene	3301.41	2.94
13.03	1,6-Octadien-3-ol, 3,7-dimethyl-	2555.99	2.10
14.97	Isopulegol	2392.28	1.81
15.36	Beta -Citronellal	7803.63	33.22
18.53	Alpha-Citronellol	5340.23	13.07
19.73	Geraniol	5960.38	21.12
23.79	Citronellyl acetate	3543.78	3.85
25.05	2,6-Octadien-1-ol, 3,7-dimethyl-, acetate	3555.17	3.92
25.47	.(-)-Beta-Elemene	2568.32	2.19
29.07	Germacrene D	2624.37	2.42
30.70	Cadinene	2836.23	1.16
31.72	Elemol	3235.42	3.77
32.71	Germacrene D-4-ol	1636.45	0.92
35.18	tau.-Muurolol	1305.89	0.92
35.63	Alpha-Cadinol	1704.98	1.25

Figure 20 GC chromatogram of citronella oil

VITA

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Publication

1. Sayowan, W., Siripornpanich V., Piriyapunyaporn T., Hongratanaworakit T., Kotchabhakdi N., and Ruangrungsi N. The effects of lavender oil inhalation on emotional states, autonomic nervous system and brain electrical activity. Journal of The Medical Association of Thailand vol. 95(4) April 2012.

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Proceeding

Sayowan, W., Siripornpanich V., Piriyapunyaporn T., Hongratanaworakit T., Kotchabhakdi N., and Ruangrungsi N. Effect of citronella oil on alpha brain wave activities. Proceeding of the 28th Annual research Conference in Pharmaceutical Sciences pp. 70-71. Bangkok, 2012.