#### **CHAPTER 3**

## Screening Thai aromatic plants by sensory acceptance, antioxidant properties and volatile characterization

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#### Abstract

The objective of this study was to investigate antioxidant properties and volatile characterization of Thai aromatic herbs and flowers. Eleven Thai aromatic plants were selected in this experiment due to its aroma characteristic and activity. All samples were dried with hot air oven at 45°C for 24 hour. Dried ground Thai aromatic plants were extracted using 70% Ethanol and evaporated using rotary vacuum evaporator. The sensory acceptance was main criteria whereas yield recovery and antioxidant activity analysis were the minor criteria. The results showed that the sensory acceptance showed that overall aroma rating from extract of kaffir lime leaves had the highest rating  $(6.6\pm0.04)$ , followed by white champaca petals  $(6.5\pm0.03)$  and champaca petals  $(6.3\pm0.03)$  without significant difference. The extract from indian cork flower gave the highest yield recovery (39.25%), followed by white champaca petals (22.83%). Ferric reducing ability power (FRAP) showed that the extract from white champaca petals gave the highest value  $(4.97\pm0.01 \text{ }\mu\text{mol trolox/g})$ . This study has shown that white champaca petals provide potential for product development using aroma and flavor based on consumer acceptance which can be further developed into food and non-food products.

**Keywords:** aromatic plant, sensory acceptance, total antioxidant activity, gas chromatography, volatile compounds characterization

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#### **3.1 Introduction**

Many scented herbs and flower provide pleasant emotion toward human perception. The aromas from herbs and flower were from volatile compounds which affected the human sensory system and have potential application as components of food and beverages (Shang, Hu, Deng, & Hu, 2002). Herb and flower have always been used for medicinal and soothing properties. Herbs commonly used as fresh or dried leaves in food cooking. Flowers have been used in beverages and sweets (El-Baz, 2006). Each herbs and flowers always have different and unique aroma with similar aroma compounds but the unique aroma of mostly came from different ratio of the mixture of volatile compounds or distinctively different volatile compound in each flower (Samakradhamrongthai, Utama-Ang, & Thakeow, 2009).

Petal and leave from Thai aromatic plants extract contained interesting aroma with some antioxidant properties. Most of affected aroma and flavor toward consumer are called dominant or character compound that mostly provided pleasant aroma which mainly originates from the petals (Linskens, Jackson, & Allen, 1997) and enhances good emotions and sensation to consumers. Moreover, it has been proved that flower aroma can stimulate blood circulation and help the respiration system (Leelapornpisid, Chansakaow, Chaiyasut, & Wongwattanakul, 2007).

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The aroma and flavor compounds are usually found in plant crude extract and volatile oil which can be secreted from plant cells, secretion ducts, and glandular hairs of plants (Calkin & Jellinek, 1994). Scented plant extract and volatile oil have been known as possessing biological activities (El-Massry, El-Ghorab, & Farouk, 2002).

They also have various pharmacological effects such as antiseptic and antispasmodic properties (Paibon *et al.*, 2011). In addition, there are others interesting possessed activities which are antibacterial, antifungal, anti-inflammatory and antioxidant properties. There are highly rising interest in the use of those volatile extract both in pharmaceutical and food industry, a systematic examination of scented plant extract for these properties has become increasingly important (El-Massry *et al.*, 2002).

In this experiment, the sensory acceptance on overall aroma of Thai aromatic herbs and flowers that provided preferable aroma toward consumer was screened by human sensory evaluation using 9-point Hedonic scaling. The antioxidant activities were analyzed using Ferric reducing ability power (FRAP) and DPPH scavenging activity. The chemical components of volatile compounds obtained from selected Thai aromatic plant were determined by gas chromatography coupled with mass spectrometer (GC-MS). The dominant volatile compounds obtained from selected Thai aromatic plant were determined by gas chromatography coupled with olfactometer (GC-O). The highest overall aroma of Thai aromatic extract with high value of antioxidant activity was further used for the next experiment.

#### **3.2 Materials and Methods**

#### 3.2.1 Herbs and flowers raw materials

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Eleven Thai aromatic plants with unique aroma which were sweet, floral and citrus that contained some bioactive activities were selected for this screening experiment as shown in Table 3.1. India cork flower, kaffir lime leaves and lemon grass was collected from Faculty of Agro-industry, Chiang mai university area during September 2011. The others were purchased from Waroroj fresh market (Chiang mai, Thailand). All samples were dried with hot air over (FD 115, Serial 08-836864, Binder, Germany) at 45°C for 24 hour (Safeena, Patil, & Naik, 2006).

Scientific name	Thai name	Common	Parts	Aromatic description	Activities	References
		Name		0101912		
Jasminum sambac L.	mali	arabian	whole	creamy, woody, floral	antidepressant, antiseptic,	Luebke (1984); Gerrard (1989); A
		jasmine	flower	0.0	antispasmodic, expectorant	Rashid (2006)
Gardenia jasminoids	pudsorn	gardenia	whole	avvect amon flored	antimizer hiel activity antioxidant	Luebke (1984); Chaichana,
			flower	sweet, green, norai		Niwatananun, Vejabhikul, Somma,
		12"	1 -		activity, antimonibolic	& Chansakaow (2009)
Millintonia hortensis L.	peeb	indian cork	whole	sweet, floral	antifungal, antimicrobial, larvicidal,	Thongpoon & Poolprasert (2014);
		flower	flower	( and	antiproliferation and antioxidant	Ramasubramaniaraja (2010)
Sesbania drummondii	sa-no	rattle box	whole	green, grassy	antibacterial, antifungal, antitumor and	Luebke (1984); Jamzad, Rostami,
		0	flower	N W/	antioxidant activity	Kazembakloo, Ghadami, &
		EI	1	A AC	$\Lambda$ / $\mathcal{S}$ //	Shafaghat (2014)
Michelia champaca L.	champa	champaca	petals	dry, floral, woody	antioxidant activity, antimicrobial, anti-	Luebke (1983); Kaiser (1989);
			1.	、月初日	inflammatory, anti-cancer activity,	P&F (2000); Geetha, Jeyaprakash,
			G.	000	ST /	& Nagaraja (2011); Ananthi &
			14	ALIMINT	3RD	Chitra (2013)
Rosa damascene Mill.	kularb	rose	petals	foral, sweet, green	antibacterial, antioxidant, antitussive,	Luebke (1983); Boskabady,
					hypnotic, antidiabetic, and relaxant	Shafei, Saberi, & Amini (2011)
	6		S 11M	o Spelo S	effect on tracheal chains	
	ด	oana	<b>u</b> n	1.19110.1d	101000100	
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Table 3.1 The aroma characteristics and biological	activities of the selected Thai aromatic	plants
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Scientific name	Thai name	Common	Parts	Aromatic description	Activities	References
		Name	1	-0181912		
Michelia alba D.C.	champee	white	petals	sweet, floral, grassy	antioxidant activity, antibacterial,	Luebke (1983); Zhu, Lu, & Xu
		champaca	20	0.0	photoaging prevention, antiasthmatic	(1982)
Cananga odorata Lamk.	kradung-nga	ylang ylang	petals	floral, sweet	antibacterial, anti-inflammatory, anti-	Luebke (1982); Taha, Arya, Ali
			`/		cancer, anti-viral, antioxidant activity	Mohd, & A. Hadi (2013)
Citrus hystrix L.	ma-krood	kaffir lime	leaves	sweet, floral, citrus	antimicrobial, antitumor,	Luebke (1984); Tangkanakul et al.,
		1 19/		( Juliuman and	antimutagenicity and antioxidant	(2009)
		305		300	activity	
Cymgopogon winterianus Jowitt	takrai	lemon grass	leaves	sweet, floral, citrus	antifungal, anticarcinogenic,	Luebke (1984); Gerrard (1989);
		JUL I		THE I	hypotensive, hypoglycemic, antioxidant	Cheel, Theoduloz, Rodriguez, &
		la		N W /	action	Schmeda-Hirschmann (2005);
		EI		A ACI	$\Lambda$ / $S$ //	Menut (2000)
Melissa officinalis L.	sa-ra-nae	Thai mint	leaves	sweet, citrus, grassy	calming, antispasmodic, strengthening	Luebke (1984); Bounihi, Hajjaj,
			12	LL SOL	heart effects, anti-inflammatory and	Alnamer, Cherrah, & Zellou
			G.	000	antioxidant activities	(2013)
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Table 3.1 (cont'd.)	The aroma characteristics	and biological activities	of the selected Th	nai aromatic plants
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#### 3.2.2 Chemical and reagents

2,2-Diphenyl-1-picrylhydrazyl (DPPH), Trolox and *n*-alkane series (C<sub>7</sub>-C<sub>20</sub>) were purchased from Sigma-Aldrich Co., LLC. (Steinheim, Germany). The 2,4,6-tris(2-pyridyl)-S-triazine (TPTZ) was purchased from Fluka Chemicals under Sigma-Aldrich Co., LLC. (Riedel de Haen, Switzerland). Acetate buffer, pH 3.6 (C<sub>2</sub>H<sub>3</sub>NaO<sub>2</sub>.3H<sub>2</sub>O) was obtained from Sigma-Aldrich Co., LLC. (Riedel de Haen, Germany). Acetic acid (C<sub>2</sub>H<sub>4</sub>O<sub>2</sub>), hydrochloric acid (HCl) and ferric chloride (FeCl<sub>3</sub>.6H<sub>2</sub>O) were obtained from BDH Laboratory Supplies (United Kingdom). Absolute ethanol and methanol were purchased from Union Science Co., Ltd. (Chiang mai, Thailand). All chemical and reagents used in this study were of analytical grade.

#### 3.2.3 Methods

#### 3.2.3.1 Herbs and flower extract preparation

Selected Thai aromatic plants were dried with hot air oven (FD 115, Serial 08836864, Binder, Germany) at 45°C for 24 hours. The moisture content of dried sample was measured in range of 8-11.5% to ensure good quality, firmness and maintain sample quality. The exceeded drying sample with moisture content lower than 8% resulted sample to be weakened adhesion and cohesion forces in plant tissue, also lead to abscission (Safeena *et al.*, 2006; Dilta, Sharma, Baweja, & Kashyap, 2011). The dried plant sample was extracted with 70% ethanol in the ratio of 1:10. The sample were shaken in solvent for 24 hr at 48 rpm (Heidolph, UNIMAX2010, Germany) and then filtered. a percentage and contained in amber vial under 4°C for further experiment (Paibon *et al.*, 2011).

# 3.2.3.2 Consumer survey and preference test on overall aroma of herbs and flower extract

The consumer survey was conducted through questionnaire querying the consumer (n=100). The personal information was consisted of gender, age, type of food that mixed aroma from scented plant. All personal data was demonstrated in term of percentage from all consumers. For the preference test, the extract of selected herbal plants were taken to evaluate using consumer acceptance (n=100) on overall aroma liking using 9-point Hedonic scale where 1 = dislike extremely, 5 = neither like nor dislike and 9 = like extremely (Resurreccion, 1998). The evaluation was taken in the booth in sensory evaluation room with Susense program (Silpakorn University, Nakorn-Pathom) at Department of product development and technology, Faculty of Agro-Industry, Chiang Mai University, Chiang mai, Thailand. The 0.05% w/v ethanolic extract solution was evaluated. The room temperature was adjusted at  $25^{\circ}$ C. Sensory evaluation of sample was evaluated three times for each treatment and each time consumer was asked to sniff coffee between each sample evaluation to cleanse the leftover of previous aroma in consumer nasal cavity (Shukla, 1997).

#### 3.2.3.3 Ferric reducing ability power (FRAP)

FRAP assay was performed by a method adapted from Benzie & Strain (1996). The FRAP reagent was prepared from 300 mmol/l acetate buffer, pH 3.6 (3.1 g Sodium acetate trihydrate) and Acetic acid 16 ml/l buffer solution. The 10 mmol/l tripyridyltriazine (TPTZ) in 40 mmol/l HCl and 20 mmol/l of Iron (III) chloride hexhydrate. Working FRAP reagent was prepared by 25 ml acetate buffer, 2.5 ml TPTZ solution, and 2.5 ml FeCl<sub>3</sub>.6H<sub>2</sub>O solution. The solution was placed at room temperature for 4 min, prior to measure the absorbance at 593 nm. Ten ml of mixed solution and sample were then added to 990  $\mu$ l of FRAP solution (ferric chloride solution: tripyridyltriazine solution:acetate buffer of 1:1:10 by volume). The absorbance was calculated by subtracting the absorbance from reagent blank. The FRAP value was calculated as  $\mu$ mol trolox/g sample using trolox standard curve.

#### **3.2.3.4 DPPH scavenging activity**

The electron donation ability of the corresponding extracts was measure from the decreasing of a purple-colored methanol solution of DPPH (Kaisoon, Siriamornpun, Weerapreeyakul, & Meeso, 2011). The molecular structure of the antioxidant and its kinetic behavior The antioxidant and radical concentration are factors that effected to be decreased (Brand-Willams, Cuvelier, & Berset, 1995). The antioxidant activity of the extracts from DPPH scavenging activity was determined according to method described in Braca *et al.* (2001) investigation. The 0.1 ml of extract was added to 2.9 ml of a 0.004% DPPH solution in methanal. The absorbance of purplecolored solution was detected at 517 nm after set the mixed solution in the dark for 30 min. The percentage of inhibition of activity was calculated as  $[A_0 - A_e / A_0 \ge 100]$  (A<sub>0</sub> = absorbance without extract;  $A_e$  = absorbance with extract).

3.2.3.5 Identification of aroma characteristics from selected herbs and flower extract using gas chromatograph olfactometer (GC-O)

Volatile compounds and aroma of extract was analyzed using gas chromatography-olfactometry (GC-O). The one microliter of 0.05% ethanolic extract was prepared sample is injected into an injection port (GC-2010, 05853, Shimadzu, Japan) coupled with an olfactometer sniffing port (O275, 1017, GL Sciences INC., Japan). The FID was operated with sampling rate at 40 msec and air flow rate of 400 ml min<sup>-1</sup>, 230°C source temperature. The GC was operated on DB-5 column (30 m x 0.53 mm, i.d., 1.50  $\mu$ m film thickness; Model 125-5032LTM , Agilent Technologies, Inc., USA), and helium is used as a carrier gas at a flow rate of 50.0 ml/min. The temperature program is started with an initial temperature of 40°C and kept for 3 min at this temperature, then heated up to 250°C at 4°C per min and held for 5 min at 250°C. 3.2.3.6 Identification of chemical constituents from volatile compound of herbal extracts using gas chromatography mass spectrometer (GC-MS)

To identified dominant volatile compounds of extract from selected plants and its characteristic aroma to indicate effect of aroma toward consumer acceptance. The 70% v/v ethanolic extract was prepared from dried Thai aromatic plants. The filtrates were evaporated under reduce pressure at the 40°C. The residue was collected for further experiment (Paibon *et al.*, 2011). Five grams of evaporated herbal extract was analyzed for volatile compounds components. The GC analysis was performed on GC-MS (QP-2010 plus system, Shimadzu, Japan) with mass-selective detector with electron impact ionization. The samples were separated using a DB-5 MS capillary column (5% phenylmethylpolysiloxane, 30 x 0.25 mm ID and 0.25  $\mu$ m film thickness) with helium as the carrier gas (0.99 ml/min). The oven temperature condition was followed GC-O method. The MS is operated in the electron impact mode with electron energy of 70 eV and scan over range 20-300 amu, 230°C source temperature. The MS interpretation was compared with WILEY 7 and the National Institute of Standards and Technology (NIST 2005, Gaithersburg, USA) as well as Linear retention indices with a series of *n-alkanes* (C<sub>7</sub>–C<sub>20</sub>).

### 3.2.4 Statistical analysis

All data were carried out in triplicate and reported as mean±standard deviation of mean (SEM). The repeated measurement analysis of variance (rm-ANOVA) was used to assess the difference of sensory rating score from preference test. Yield recovery and antioxidant activity were analyzed using analysis of variance (ANOVA) using the Duncan's multiple range test (DMRT). The statistical analysis was conducted using SPSS 17.0 (SPSS Inc., IBM Corp., Chicago, IL, USA) with significant level at 95% (p < 0.05).

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#### 3.3 Results and discussion

#### 3.3.1 Consumer survey and aroma preference test

One hundred and six consumers participated in consumer survey which were 50 male (47.2%) and 56 female (52.8%). The age range of consumers was divided into six groups: less than 15 years old, 16 - 25 years old, 26 - 35 years old, 36 - 45 years old, 46-55 years old, and more than 55 years old. The most of the consumers were in among age range of 26 - 35 years old (19.8%). The general information of 106 consumers was shown in Table 3.2. The result from consumer survey showed that the most preferable aroma from consumer opinion can be mix with sweets at the highest percentage (89.6%), followed by bakery product (22.6%), beverages product (15.1%), and food product (2.8%) (Table 3.3). The consumer survey also suggested that fresh dessert was the most acceptable idea to develop product with herb/flower aroma at the highest percentage (84.9%), followed by frozen dessert (60.4%), beverages product (34.0%) fresh bakery product (8.5%), frozen bakery product (8.5%), and fresh food product (2.8%) (Table 3.4). For the preference test, the rm-ANOVA for the overall aroma liking show wilks' lambda value toward 1.0 which suggested non-significant value among repeated measurement as discussed in Seltman (2015). The result can be implied that the time of evaluation of all selected aromatic plant was not significant as shown in Table 3.5. The preference test on overall aroma liking showed significant difference. Kaffir lime leaves was rated the highest (6.6±0.04), followed by white champaca petals  $(6.5\pm0.03)$  and champaca petals  $(6.3\pm0.03)$  (Table 3.6). The high and medium intensity of main aroma attributes were major factor affected overall aroma rating (Samakradhamrongthai, 2011). The preference test demonstrated significant difference similar characteristic notes of kaffir lime leaves, white champaca petals, and champaca petals but there was variation of volatile compound composition that provided disparity of those samples resulted from the imparity of overall aroma rating.

	Consumer Data	Frequency	Percentage (%)
gender	male	50	47.2
	female	56	52.8
		106	100.0
age	less than 15 years old	17	16.0
	16-25 years old	17	16.0
	26 -35 years old	21	19.8
	36-45 years old	16	15.1
	46-55 years old	16	15.1
	more than 56 years old	19	18.0
	0,00	106	100.0

**Table 3.2** General information of consumer survey (n=106)

**Table 3.3** The consumer survey on product with the most preferable aroma in consumer opinion

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Product	Frequency	Percentage (%)	
Food product	3	2.80	
Bakery product	24	22.60	
Sweets product	95	89.60	
Beverages product	16	15.10	

Note: consumer can make decision on multiple selections.

**Table 3.4** The consumer survey on product development with the most preferable aroma in consumer opinion

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Frequency	Percentage (%)
3	2.80
-	-
90	84.90
64	60.40
9	8.50
9	8.50
36	34.00
	Frequency           3           -           90           64           9           9           3

Note: consumer can make decision on multiple selections.

Table	3.5	Repeated	measurement	analysis	of	variance	from	selected	Thai	aromatic
plants										

Thai aromatic plants	Common Name	Part	Wilks'	F-value	Sig.
			Lambda		$(p \le 0.05)$
Jasminum sambac L.	arabian jasmine	whole flower	0.984	1.735	0.191
Gardenia jasminoids	gardenia	whole flower	0.976	1.297	0.278
Millintonia hortensis L.	indian cork flower	whole flower	0.958	2.363	0.105
Sesbania drummondii	rattle box	whole flower	0.991	0.960	0.329
Michelia champaca L.	champaca	petals	0.983	1.791	0.184
Rosa damascene Mill.	rose	petals	0.996	0.227	0.797
Michelia alba DC.	white champaca	petals	0.994	0.674	0.414
Cananga odorata Lamk.	ylang ylang	petals	0.975	2.673	0.105
Cymgopogon winterianus	lemon grass	leaves	0.991	1.000	0.320
Jowitt	1 Louis		71-		
Citrus hystrix L.	kaffir lime	leaves	0.969	3.311	0.072
Melissa officinalis L.	Thai mint	leaves	0.994	0.598	0.441

**Table 3.6** Overall aroma rating from selected Thai aromatic plants

Thai aromatic plants	Common Name	Part	Overall aroma liking					
Jasminum sambac L.	arabian jasmine	whole flower	4.7±0.04g					
Gardenia jasminoids	gardenia	whole flower	4.7±0.04fg					
Millintonia hortensis L.	indian cork flower	whole flower	4.4±0.05h					
Sesbania drummondii	rattle box	whole flower	4.1±0.03j					
Michelia champaca L.	champaca	petals	6.3±0.03c					
Rosa damascene Mill.	by rose lang	petals	4.3±0.02i					
Michelia alba DC.	white champaca	petals	6.5±0.03b					
Cananga odorata Lamk.	ylang ylang	petals	$4.8 \pm 0.07 f$					
Cymgopogon winterianus Jowitt	lemon grass	leaves	5.4±0.01d					
Citrus hystrix L.	kaffir lime	leaves	6.6±0.04a					
Melissa officinalis L.	Thai mint	leaves	5.2±0.02e					

*Note:* The different letters in the same column mean significant difference ( $p \le 0.05$ )

#### **3.3.2 Extract yield recovery**

The physical appearance of the extract were found to be greenish and yellowish to pinkish-orange as conformed to Leelapornpisid et al.(2007), which suggested that extract physical appearance showed range of color from green, light yellow, brown, pink and light orange. Yield recovery of eleven Thai aromatic plants from Indian cork flower was the highest (39.25%) followed by white champaca petals (22.83%) and damask rose petals (18.21%), respectively (Table 3.7). The yield recovery from Indian cork flower was higher than cold enfleurage, hot enfleurage and petroleum ether extraction as shown in research from Paibon et al. (2010) as well as yield recovery of white champaca petals that higher than hexane and petroleum ether extraction (Punjee et al., 2009). In contrast, ethanolic of damask rose petals showed lower yield recovery when applied 70% v/v ethanol on extraction. The reason affected yield recovery when using ethanolic extraction in this experiment was polarity of solvent and extract. The extract yield depended on the ability of the solvent to extract volatile compounds with various polarities (Rydberg, Musikas, & Choppin 1992; Calkin & Jellinek, 1994). The result showed that Indian cork flower and white champaca petals had more polar molecules which indicated that there were more polar active molecules such as phenolic acid derivatives and flavonoid compounds as suggested in many investigations that higher yield recovery of extract from some plants contained high level of phenolic compound and antioxidants properties (Borneley & Peyrat-Maillard, 2000; Paibon et al., 2010). เหาวทยาลยเ

# 3.3.3 Total antioxidant activity using Ferric reducing ability power assay (FRAP)

The reducing ability of a compound mostly depends on the presence of reductants which have been exhibited antioxidant potential using breaking free radical chain reaction (Naik *et al.*, 2003). The results showed that FRAP value from Thai aromatic plants extract solutions were ranging from 1.290–4.971 µmol trolox/g sample. White champaca petals exhibited highest reducing ability power ( $4.971\pm0.015$  µmol

trolox/g sample) followed by ylang ylang petals ( $4.526\pm0.01 \mu$ mol trolox/g sample) and indian cork flower ( $4.526\pm0.01 \mu$ mol trolox/g sample) (Table 3.7). The phenolic hydroxyl group presented in plants extract caused the difference of FRAP value (Kaisoon *et al.*, 2011).

#### 3.3.4 Free radical scavenging activity using DPPH assay

Free radical scavenging activity of extract was ranging from 56.88–83.68%. DPPH scavenging activity from damask rose petals was the highest (83.68%) (Table 3.7). However, the DPPH scavenging activity from others was not reached 80% as expected but kaffir lime leaves and white champaca petals taken to consider due to high rating of consumer acceptance. The DPPH scavenging activity of kaffir lime leaves and white champaca petals were 70.32% and 72.12% which exhibited high antioxidant activity as well (Leelapornpisid *et al.*, 2007; Hasan *et al.*, 2009). From Table 3.7, the scented plants with high yield recovery also exhibited high antioxidant activity on account of high amount of phenolic compounds as stated in the studied on antioxidant activity in selected plants of Wong *et al.* (2006). The yield recovery was ranked from indian cork flower (39.25%), white champaca (22.83%), damask rose (18.21%) and jasmine (11.60%), respectively which also showed high antioxidant activity as white champaca, indian cork flower, damask rose, respectively.

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Thai aromatic plants	Common Name	Part	Yield recovery	FRAP	DPPH
	- N	110	(%w/w)	(µmol trolox/g sample)	(% Scavenging)
Jasminum sambac L.	arabian jasmine	whole flower	11.60±0.04d	3.652±0.016e	56.88±0.46g
Gardenia jasminoids	gardenia	whole flower	4.53±0.09h	1.290±0.001h	76.98±0.99bc
Millintonia hortensis L.	indian cork flower	whole flower	39.25±0.12a	4.526±0.011b	73.12±1.58d
Sesbania drummondii	rattle box	whole flower	0.53±0.17j	3.831±0.009d	82.74±0.69a
Michelia champaca L.	champaca	petals	10.70±0.04e	4.508±0.042c	77.95±1.35b
Rosa damascene Mill.	rose	petals	18.21±0.04c	1.290±0.002h	83.68±2.22a
Michelia alba DC.	white champaca	petals	22.83±0.13b	4.971±0.015a	72.12±0.67de
Cananga odorata Lamk.	ylang ylang	petals	11.09±0.17e	4.526±0.011b	75.65±0.65c
Cymgopogon winterianus Jowitt	lemon grass	leaves	5.30±0.10g	2.103±0.010g	75.93±1.10c
Citrus hystrix L.	kaffir lime	leaves	2.07±0.01i	1.136±0.005i	70.32±0.37e
Melissa officinalis L.	kitchen mint	leaves	9.26±0.03f	2.923±0.007f	66.28±074f

Table 3.7 Yield recoveries and antioxidant activity from selected Thai aromatic plants

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## **3.3.5 Identification of aroma characteristics from selected herbs and flower** extract using gas chromatograph olfactometer (GC-O)

Terminology of aroma from three scented plant extract was discussed and agreed upon panelists. Those aromatic solutions had similar characteristic note which were citrus, floral and sweet. Kaffir lime leaves provided high intensity of citrus-like characteristic and medium intensity of floral characteristic whereas white champaca petals and champaca petals released medium intensity of floral characteristic (Table 3.8). Those characteristic notes were affected toward consumer acceptance because of its pleasant aroma which indicated in Castro-Vazquez, Diaz-Moroto, Gonzalez-Vinas, & Perez-Coello (2009) research that trained panels most likely to rate higher score for sample that contained aroma associated with citrus, floral, aromatic herbs fresh fruit and ripe fruit, respectively. Even though, characteristic aromas showed the aroma resemblance but there were differences of volatile compound and its variation of concentration which provided its unique characteristic (Surburg, Guentert, & Harder, 1993). This suggested that higher rating score of consumer acceptance from citrus aroma followed by floral aroma and aromatic herbs aroma. Kaffir lime leaves, champaca petals, and white champaca petals were selected because of high rating in overall aroma to identified dominant characteristic aroma and volatile compounds. The dominant characteristic odors of kaffir lime leaves, champaca petals, and white champaca petals were different as shown in Table 3.9. Highly trained panel sniffed for main characteristic aroma from extract solution of kaffir lime leaves, champaca petals, and white champaca petals. Key compounds were identified using direct intensity which agreed upon panel's opinion as well as terminology of those key compounds. There were three levels of intensity which were weak, medium and strong. Main aroma from extract characteristic odor of citrus and sweet (kaffir lime leaves), champaca flower /incent-like and honey (champaca petals), and white champaca flower (white champaca petals).

Term of	Definition	References
aroma		
Citrus-like	aromatic associated with citrus fruits, leaves and peel	Lemon zest
Floral	aromatic associated with materials that have a sweet	Mixed Michelia
	aroma with relation of flower scent in general	sp. petal
Incent	aromatic associated with materials that related to	Ground incent
	sandal wood and incent aroma	
Sweet	aromatic associated with materials that have a sweet	Tangerine
	aroma and related to flower scent	
Honey	aromatic associated with materials that related to	Honey
•	spics and savory aroma	-

Table 3.8 Term and definition and reference sample of character aromas

Table 3.9 Characteristic odors of selected Thai aromatic plants extract using GC-O



\*C = Citrus-like, FL = Floral, IC = Incent, SW = Sweet, HN = Honey

# **3.3.6 Identification of chemical constituents from volatile compound of herbal extracts using gas chromatography mass spectrometer (GC-MS)**

From Table 3.10, the identified volatile compound from kaffir lime leaves showed high amount of menthoglycol, followed by ethyl hexadecanoate, citronellal, and 2-nonen-4-one. The identified volatile compounds from champaca petals showed high amount of beta-elemene, followed by beta-selinene, trans-caryophyllene, alphaselinene, and delta-cadinene and the identified volatile compounds from white champaca petals showed high amount of 2-methyl butyric acid, followed by linalool and verbenone.

Moreover, the characteristic aromas from GC-O were acknowledged together with GC-MS. The characteristic aroma and volatile compounds of three selected aromatic plants were identified as menthoglycol (kaffir lime leaves), beta-elemene and trans-caryophyllene (champaca petals) and 2-methyl-butanoic acid and linalool (white champaca petals). In addition, the identified characteristic aromas were demonstrated antioxidant activity and antibacterial activity (Miguel, 2010; Prag, 2012; Guleria *et al.*, 2013). In conclusion, the selected aromatic plants extract showed characteristic aromas that affected consumer acceptance in high rating, also provided potential used in food product as flavoring agent that contained high antioxidant activity.

The identified characteristic volatile compounds of three selected Thai aromatic plants were menthoglycol (kaffir lime leaves), beta-elemene and trans-caryophyllene (champaca petals) (Samakradhamrongthai, 2011), and 2-methyl-butanoic acid and linalool (white champaca petals) (Pensuk, Padumanond, & Pichaensoonthon, 2007). In addition, the main characteristic volatile compounds of three selected Thai aromatic plant also demonstrated antioxidant activity. Menthoglycol from kaffir lime leaves showed antioxidant activity and antibacterial activity with insect repellants effect (Prag, 2012). Beta-elemene and trans-caryophyllene from champaca exhibited antioxidant, anti-inflammatory and antibacterial activities according to a review from Miguel (2010) and addition research from Medeiros *et al.* (2007) and Fernandes *et al.* (2007). Linalool

from white champaca also exhibited antioxidant activity (Guleria *et al.*, 2013), antibacterial and antifungal activity as the findings from Bougatsos, Ngassapa, Runyoro, & Chinou (2004) and Cha, Jung, & Lee (2007) researches.

 Table 3.10 Identified volatile compounds from Kaffir lime, Champaca and white

 champaca using gas chromatograph mass spectrometry

	Compounds	LRI <sup>[a]</sup>	LRI <sup>[b]</sup>	Characteristic odor <sup>[c]</sup>
		(Stu. arkane solution)	(WILET/)	
	Kaffir lime leaves	, , , , , , , , , , , , , , , , , , , ,	200	91
1	citronellal	1192	1153	sweet/dry herbal/citrus
2	beta-citronellol	1249	1231	sweet/waxy/citrus
3	menthoglycol	1248	1301	herbal/minty
4	nerolidol	1586	1534	waxy/floral
5	elemol	1518	1557	sweet/woody/green
6	torreyol	1525	1644	herbal/astringent/sweet
7	ethyl hexadecanoate	1934	2013	waxy/creamy/hint of balsam
8	ethyl linoleate	2014	2055	mild/fatty/fruity
	Champaca	1 8	6	
1	phenyl ethyl alcohol	1053	1060	floral
2	linalool oxide	1102	1076	floral/green
3	2-methoxy-4-vinyl phenol	1222	1131	spice/peppery/woody
4	methyl anthranilate	1249	1346	sweet/fruity
5	alpha-copaene	1274	1372	woody/earthy
6	beta-elemene	1285	1392	waxy/herbal
7	beta-santalene	1409	1421	woody/earthy
8	trans-caryophyllene	1411	1429	sweet/woody/citrus
9	alpha-bergamotene	1420	1436	warm/tea leaf like
10	alpha-humulene	1442	1451	woody/earthy
11	alpha-curcumene	1461	1475	herbal
12	beta-bisabolene	1482	1515	herbal
13	(-)-caryophyllene oxide	1548	1580	sweet/woody
14	spathulenol	1619	1619	earthy/herbal/fruity
15	ethyl hexadecanoate	1934	1933	waxy/creamy
16	methyl linoleate	1956	2013	bland
17	ethyl linoleate	2093	2055	mild/fatty/fruity
	White champaca			
1	2-methyl butyric acid	892	858	sweet/ripen fruit
2	terpinolene	1092	1083	sweet/piney/citrus
3	diethyl malonate	0 1111 C	1069	sweet/green apple
4	hotrienol	1132	1101	sweet/spicy
5	phenyl ethyl alcohol	1141	1139	floral
6	lilac aldehyde	1153	1256	floral/lilac
7	linalool	1218	1277	floral/fruity
8	decanal	1230	1294	sweet/waxy/citrus
9	terpendiol	1290	1343	piney
10	verbenone	1348	1344	minty/spicy
11	menthoglycol	1391	1391	herbal/minty
12	alpha-calacorene	1512	1538	dry-woody
13	spathulenol	1619	1619	earthy/herbal/fruity
14	farnesyl acetate	1746	1820	green/floral/rose
15	ethyl hexadecanoate	1873	2013	waxy/creamy
16	ethyl linoleate	2008	2055	mild/fatty/fruity

 $LRI^{[a]} = Kovats$  retention index relative standard alkane series  $(C_7 - C_{20})$ .

LRI<sup>[b]</sup> = Kovats retention index from WILEY 7 and the National Institute of Standards and Technology (NIST 2005)

[c] Characteristic odor of identified volatile compounds from www.pherobase.com and www.flavornet.org.

#### 3.4 Conclusion

These results suggested that Thai aromatic plants extract had unique and interesting aroma. The consumer survey showed that the most preferable aroma from selected plant can be applied to dessert product. The preference test showed that kaffir lime leaves, champaca petals, and white champaca petals were rated higher than others. Yield recovery of indian cork flower was the highest followed by white champaca petals whereas white champaca petals exhibited high rate of reducing ability power and DPPH scavenging activity. Selected plants extract were provided dominant characteristic odor of citrus and sweet (kaffir lime leaves), champaca flower /incent-like and honey (champaca petals), and White champaca flower (white champaca petals). The characters that affected consumer acceptance were Citrus, Flower, and Sweet. Thrityfour volatile compounds were identified in total. Kaffir lime leaves showed high amount of menthoglycol whereas champaca petals showed high amount of beta-elemene and trans-caryophyllene which were different from white champaca petals that showed high amount of 2-methyl butyric acid and linalool. The main characteristic volatile compounds of three selected Thai aromatic plant which were menthoglycol, betaelemene, trans-caryophyllene, and linalool also demonstrated antioxidant activity. This study has shown differences between Thai aromatic plants extract in consumer acceptance and perspective of some antioxidant activity with characteristic odor and its volatile compounds. The results tended to show that white champaca petals provide potential for product development using aroma and flavor based on consumer acceptance with antioxidant activity which can be further developed into food and nonby Chiang Mai University food products. rights reserved

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