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# Volatile Compounds of a Traditional Thai Rice Wine

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**Abstract:** The three best sellers of Thai rice wines, Sato, from the Northeast of Thailand were collected to analyze volatile composition of the products. The contents of principal volatile compounds were determined using gas chromatography-flame ionization detection. The results showed that there were at least 27 volatile compounds detected at various concentrations in the three samples. To achieve the identification of major would be impact odorants of Sato, the aroma index was calculated. From the 27 compounds identified, 10 compounds were determined to be the most powerful odorants (aroma index more than 1): acetaldehyde, ethyl acetate, ethyl butyrate, ethyl decanoate, 2-phenethyl acetate, isobutyl alcohol, isoamyl alcohol, 1-hexanol, isovaleic acid and caproic acid.

**Key words:** Aroma index, rice wine, volatile compounds

## INTRODUCTION

As rice wines contain rich nutrients, they are excellent for bringing up a good appetite and good health upon regular drinking in suitable amounts. They can also be used as a cooking additive to relieve stinking and make dishes more delicious. In ancient times, the rice wines were considered as the best of all medicines. A large amount of healthy alcoholic drinks is based on rice wines.

Sato, a Thai rice wine, is a traditional alcoholic beverage in Thailand, which contains ethanol not higher than 15% (by volume). Sato has been the most popular alcoholic drink for Thais since ancient times. In general, it is manufactured at home or by small cottage industries. Sato is made from white or black glutinous rice using Loog-Pang or Chinese yeast cake as a dry-starter. Sato production process (Fig. 1) is similar to the production process of Balinase rice wine (Sujaya et al., 2004) and Vietnamese rice wine (Dung et al., 2007). Initially, polished glutinous rice is washed, steeped and steamed. After being cooled to room temperature, the rice is inoculated with starter powder. The fermentation is carried out in an earthen jar at room temperature for 5 days. The mixture is then added with clean water and fermented for 1 month. Lastly, Sato is filtered and pasteurized before packaging.

Loog-Pang is a microbial inoculum that is stored in dried form for traditional Thai rice wine starter. It is mixed with starch, boiled and cooled water, microorganisms and

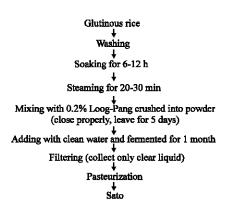


Fig. 1: Flow charts of Sato production process (Rittiplang, 2006)

herbs or spices. Microorganisms responsible for effecting important changes in the Sato production are fungi, yeast and lactic acid bacteria. Loog-Pang has been made in Thailand from family to family for centuries. Normally, the recipes are kept secret.

Similar traditional rice wines have equivalent appellations, for example as Japanese Sake and Mirin, Vietnamese Ruou, Philippine Tapuy, Indonesian Brem, Malaysian Tapai, Korean Makkulli and Yakju and Chinese Huang jiu, Huadiao jiu, Shaohsing, Chia Fan, Hsiang Hsueh, Shan Niang and Yen Hung.

The aroma of wines is the product of a biochemical and technological sequence. It is important for wine as it makes a major contribution to the quality of final product (Asano et al., 1999; Torrea et al., 2003; Selli et al., 2004; Yongmei et al., 2007). The aroma of Sato depends on two main factors; environmental and management practices such as rice varieties, type of Loog-Pang, water, Satomaking techniques etc. There are some evidences indicating that compounds i.e., alcohols, acids, aldehydes and esters play an important role in the flavors and sensory properties of alcoholic beverages (Soufleros et al., 2004; Apostolopoulou et al., 2005; Selli et al., 2006). Most of them appear during fermentation process and their concentrations vary over a wide range. However, the volatile composition of Sato has not been reported.

This research aims to identify and qualify principal volatile substances of the Sato using a gas chromatographic method. Potential aroma compounds in the Sato on the basis of aroma index (I) or odor activity values (OAV) were determined. The principle volatiles detected were also compared with those of other alcoholic beverages.

## MATERIALS AND METHODS

**Samples:** The three best sellers of Sato year 2005 were collected from leader supermarkets in Thailand and transported to the Biotechnology Laboratory at the Department of Biotechnology, Faculty of Technology, Khon Kaen University, Thailand for analysis.

A 100 mL portion of the samples was transferred into a 500 mL round bottle and added with a 35 mL of deionized water. The mixture was distilled by using a distillation apparatus (Zoecklein *et al.*, 1990). The distillate was collected to approximately 95 mL on ice bath and then it was made up to 100 mL with deionized water. In order to avoid the loss of volatile compounds, the mixture was stored in a glass screw-top vial at -20°C before further analysis.

Reagents: Ethyl butyrate, ethyl decanoate, ethyl L-lactate, isobutyl acetate, diethyl succinate, ethyl laurate, ethyl caprylate, propyl acetate, ethyl caproate, hexyl acetate, 2-phenethyl acetate, 1-hexanol, 4-penten-1-ol, phenethyl alcohol, 4-methyl-1-pentanol, 3-methyl-1-pentanol, isovaleic acid, caproic acid, 2-furaldehyde and 1-pentanol were purchased from Fluka Company (St. Gallen, Switzerland). All reagents used were GC-grade quality. Acetaldehyde, 1-butanol, ethanol and ethyl acetate used were HPLC-grade quality and were purchased from BDH Laboratory Supplies (Poole, UK). Methanol, propanol,

isobutyl alcohol and isoamyl alcohol were analyze-grade quality and were purchased from BDH Laboratory Supplies (Poole, UK).

Gas Chromatographic (GC) analysis: GC analysis of volatile compounds in the distillated sample was performed using a GC-17A (Shimadzu) equipped with a fused capillary column coated with ZB-Wax (60 m× 0.25 mm diameter, 0.25 µm film thickness, Phenomenex, USA). The flow rate of carrier gas (helium) was 2 mL min<sup>-1</sup>. One microliter of the sample was injected with split/splitless mode (split ratio 1/10) by direct injection into a GC. One-pentanol was used as an internal standard. The injector temperature was maintained at 150°C. The oven temperature was programmed at 55°C for 6 min, from 55 to 120°C at 8°C min<sup>-1</sup>, held 10 min at 120°C, from 120 to 150°C at 10°C min<sup>-1</sup>, then held 18 min at 150°C. The flame ionization detector (FID) was set at 160°C. Analysis was carried out in triplicate and all data were presented as mean±standard deviations (SD).

## RESULTS AND DISCUSSION

The chromatogram of the standard volatile compounds using GC was shown in Fig. 2 while the concentrations of the analyzed volatile compounds of Sato were shown in Table 1. The results showed that there were at least 27 volatile compounds detected in the Sato samples. The samples exhibited ethanol contents ranging from 9.08 to 10.48% (by volume) which did not exceed the maximum acceptable level (15%, by volume) of fruit wines and Sato. These concentrations were not significantly different among the samples. Ethanol concentration of Sato samples depended on strains of microorganisms in Loog-Pang which directly involved in Sato production. Alcohols and esters were shown to be the largest groups of the volatile compounds in the Sato samples. These compounds are mainly produced during alcoholic fermentation (Selli et al., 2006). The alcohols occur in varying amounts and they can be recognized by their strong and pungent smell and taste (Rocha et al., 2004). Torrea et al. (2003) reported that among volatile compounds, esters had a crucial importance as they provided pleasant aroma sensation.

Acetaldehyde concentration of Sato number 3 (158.81 mg  $L^{-1}$ ) was 5-12 times higher than those of Sato numbers 1 and 2. However, it was lower than that of aging sherry fino wines (254-257 mg  $L^{-1}$ ) (Moreno *et al.*, 2005). The acetaldehyde concentrations of Sato numbers 1 and 2 (12.42 and 30.88 mg  $L^{-1}$ , respectively) were similar to that of sake (16-25 mg  $L^{-1}$ ) (Asano *et al.*, 1999). Acetaldehyde is originated from fermentation raw

## Biotechnology 7 (3): 505-513, 2008

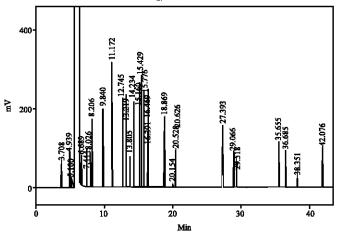


Fig. 2: Typical chromatogram of the standard volatile compounds at the optimum chromatographic conditions (3.708, acetaldehyde; 4.939, ethyl acetate; 5.100, methanol; 6.689, propyl acetate; 7.444, isobutyl acetate; 8.026, ethyl butyrate; 8.206, propanol; 9.840, isobutyl alcohol; 11.172, butanol; 12.745, isoamyl alcohol; 13.219, ethyl caproate; 13.805, pentanol; 14.234, hexyl acetate; 15.160, 4-pentene-1-ol; 15.429, 3-methyl-1-pentanol; 15.776, 4-methyl-1-pentanol; 16.396, 1-hexanol; 16.469, ethyl L-lactate; 18.869, ethyl capryrate; 20.054, unknown; 20.528, furfural; 20.626, unknown; 27.393, ethyl decanoate; 29.066, isovaleic acid; 29.518, diethyl succinate; 35.655, 2-phenethyl acetate; 36.685, ethyl laulate; 38.351, caproic acid; 42.076, phenethyl alcohol)

Table 1: Major volatile composition in the three best sellers of Sato from leader supermarkets in Thailand

	Concentrations (n	ng L <sup>-1</sup> )		Odor threshold	
Compounds	Sato 1	Sato 2	Sato 3	(mg L <sup>-1</sup> )*	Odor description*
Esters, acetates and					·
aldehydes (mg L <sup>-1</sup> )					
Acetaldehyde	12.42	30.88	158.81	110	Pungent, ripe apple
Ethyl acetate	255.61	15.35	567.43	12	Pineapple, vamish, balsamic
Propyl acetate	2.27	1.71	3.87	4.7	Celery
Isobutyl acetate	ND	ND	0.17	1.6	Sweet, fruity, apple, banana
Ethyl butyrate	32.20	65.50	82.72	0.4	Strawberry, apple, banana
Hexyl acetate	ND	0.22	ND	0.67	Apple, cherry, pear, floral
Isopentyl acetate	ND	ND	0.45	150	Buttery, cream, sweet, fruity
Ethyl-L-lactate	ND	ND	ND	0.58	Sweet, floral, fruity, banana, pear, brandy
Ethyl capryrate	ND	ND	ND	0.5	Brandy, fruity, grape
Ethyl decanoate	1.07	1.46	0.66	1200	Fruity, wine
Diethyl succinate	9.75	2.88	3.92	1.8	Fruity
2-Phenethyl acetate	3.47	0.37	0.31	0.08	Fruity, green apple, banana, wine-like
Ethyl caproate	ND	ND	ND		Sweaty, waxy, floral, soapy
Ethyl laurate	1.45	0.40	ND		
Sum of esters, acetates	318.24	118.78	818.33		
and aldehydes (mg $L^{-1}$ )					
Other alcohols (mg $L^{-1}$ )				500	Alcohol
Methanol	5.15	0.96	1.66	306	Ripe fruit, alcohol
1-Propanol	ND	ND	ND	150	Medicinal, phenolic
1-Butanol	0.50	1.29	10.21	75	Alcohol, solvent
Isobutyl alcohol	75.79	60.01	77.76	60	Nail polish, alcohol
Isoamyl alcohol	79.06	98.99	155.90		
4-Penten-1-ol	1.29	6.07	58.33	50	
4-Methyl-1-pentanol	0.01	ND	ND	50	Pungent, solvent, green
3-Methyl-1-pentanol	22.33	0.10	0.06	1.1	Herbaceous, wood
1-Hexanol	217.55	42.28	106.75	200	Rose
Phenethyl alcohol	31.14	23.08	30.30		
Sum of alcohols (mg L <sup>-1</sup> )	432.81	232.78	440.96		
Ratio sum of esters, acetates	0.74	0.51	1.86		
and aldehydes to alcohols					
Acids (mg L <sup>-1</sup> )					
Isovarelic acid	5.04	ND	ND	0.25	Fatty-rancid, cheesy
Caproic acid	3.94	0.22	ND	3	Rancid, cheese, fatty
Others (mg L <sup>-1</sup> )					
2-Furaldehyde	3.69	ND	ND	15	Sweet, almond

ND: Not Detectable, \*Peinado et al. (2004), Rocha et al. (2004), Moreno et al. (2005) and Peinado et al. (2006)

materials. It is considered to be the result of spontaneous or microbial mediated oxidation (Soufleros et al., 2004). Sensory descriptors for acetaldehyde range from classic, nutty and sherry-like to being reminiscent of overripe bruised apples (Fugelsang, 1997). Acetaldehyde levels in wines and distillates also increase during aging due to chemical oxidation of ethanol. Further oxidation of acetaldehyde may result in formation of small amounts of acetic acid (De LA Presa-owen and Noble, 1997). The reaction of ascorbic acid and oxygen may also generate hydrogen peroxide which can react with ethanol, reducing in acetaldehyde formation (Zoecklein et al., 1995). Acetaldehyde is however decreased because it can interact with ethanol resulting in the production of acetal (Mangas et al., 1996). Zoecklein et al. (1995) reported generally had acetaldehyde wines concentrations of less than 75 mg L<sup>-1</sup> after fermentation. The sensory threshold of acetaldehyde ranges of 100-125 mg L<sup>-1</sup>, therefore, the higher concentration in Sato number 3 may impart an odor of the drink.

Ethyl acetate is a main ester occurring in wines and has a significant effect on the organoleptic characteristics of wines and distillates. It is described as having fingernail polish remover properties. Ethyl acetate at concentrations of 150-200 mg L<sup>-1</sup> can add spoilage notes to the wines. At lower concentrations, ethyl acetate contribute to fruity properties of wine (Apostolopoulou et al., 2005; Zoecklein et al., 1995). Increased ethyl acetate concentrations are indicative of prolonged storage of the raw material and probable acetic bacterial spoilage. Ethyl acetate concentration in the studied samples quite varied ranging between 15.35 and 567.43 mg L<sup>-1</sup>. It was present in higher contents in Sato numbers 1 and 3, respectively. The results implied that the two samples were probably contaminated by acetic acid bacteria during the Sato fermentation. In addition, high ethyl acetate concentration in Sato numbers 1 and 3 could add spoilage flavor. In this study, ethyl acetate concentrations were however similar to those of Portuguese Bagaceiras (50.4-528 mg L<sup>-1</sup>) (Silva et al., 1996) and Chenin blanc wines (32.0-724.2 mg L<sup>-1</sup>) (Lilly et al., 2000) whereas ethyl acetate levels in sake were in the range of 115-183 mg  $L^{-1}$  (Asano *et al.*, 1999). Low ethyl acetate concentration in Sato number 2 suggested that it contribute to fruity properties of Sato.

Propyl acetate and isobutyl acetate are described as having celery and sweet, fruity, apple and banana to wines respectively (Peinado *et al.*, 2004, 2006). Propyl acetate concentrations of the Sato samples were in the range of 1.71-3.87 mg L<sup>-1</sup> which were significantly higher than those of sherry fino wines (0.04-0.33 mg L<sup>-1</sup>)

(Moreno *et al.*, 2005). Isobutyl acetate concentration of the Sato samples was low ranging from ND-0.17 mg  $L^{-1}$ . However, it was lower than that of sake (0.33-1.27 mg  $L^{-1}$ ) (Inoue *et al.*, 1994) and Monastrell wines (0.27-1.10 mg  $L^{-1}$ ) (Moreno *et al.*, 2005). The results suggested that isobutyl acetate concentrations of the Sato samples in this study have no negative effect on the Sato quality.

Ethyl butyrate (ethyl butanoate) is a higher ester contributed to wine odor (Selli *et al.*, 2006). It is described as having strawberry, apple and banana to wines (Peinado *et al.*, 2006). Ethyl butyrate levels of this study were in the range of 32.20-82.72 mg L<sup>-1</sup> which were higher than those found in kiwifruit wines (0.01-0.27 mg L<sup>-1</sup>) (Soufleros *et al.*, 2001), Chenin blanc wines (0.5-0.6 mg L<sup>-1</sup>) (Lilly *et al.*, 2000) and sherry fino wines (0.16-1.49 mg L<sup>-1</sup>) (Moreno *et al.*, 2005).

Ethyl caproate (ethyl hexanoate) is an ester contributed to wine odor (apple-like aroma) (Lilly *et al.*, 2000). It is especially important for a pleasant fruity note and it is a factor showing the sake quality. Ethyl caproate contents of sake, wines and Chenin blanc wines were 0.03-0.37, 0.16 and 1.4-14.1 mg L<sup>-1</sup>, respectively (Lilly *et al.*, 2000; Inoue *et al.*, 1994; Soufleros *et al.*, 2001). The ethyl caproate concentrations of the Sato samples were however not detected in this study.

Hexyl acetate, acetic acid ester, is characterized by aroma of flowers and fruits at low contents. It was found in the range of 0.03-0.04 mg L<sup>-1</sup> in kiwifruit wines (Soufleros *et al.*, 2001), 0.06-0.65 mg L<sup>-1</sup> in Potuguese Bagaceiras (Silva *et al.*, 1996) and 0.5-5.0 mg L<sup>-1</sup> in Chenin blanc wines (Lilly *et al.*, 2000). In this study, hexyl acetate was only detected in Sato number 2 at the concentration of 0.22 mg L<sup>-1</sup>.

Ethyl-L-lactate is considered to stabilize the distillate flavor and to soften the harsh flavor characteristics when it is present in low concentrations. It is also described as having buttery, cream, sweet and fruity to wines (Peinado *et al.*, 2006). Lactic acid bacteria spoilage increases its concentration and negatively contributes to the distillate organoleptic quality (Apostolopoulou *et al.*, 2005). It has been reported that ethyl-L-lactate production was mainly due to malolactic fermentation (Selli *et al.*, 2006). Ethyl-L-lactate of kiwifruit wines, Chenin blanc wines and sherry fino wines were in the range of 0.01-5.85, 1.2-8.4 and 81.6-471 mg L<sup>-1</sup>, respectively (Moreno *et al.*, 2005; Lilly *et al.*, 2000; Soufleros *et al.*, 2001). In this study, it was however not detected.

2-Phenethyl acetate is described as a fruity and flowery flavor with a honey note (Lilly *et al.*, 2000). Maicas *et al.* (1999) depicted that after malolactic fermentation, 2-phenethyl acetate contents of red wines

showed more significant increments. They suggested its beneficial contribution to the wine's final aroma. 2-Phenethyl acetate concentrations of the red wines were lower than those of the Sato samples (0.31-3.47 mg L<sup>-1</sup>). In this study 2-phenethyl acetate contents were similar to those of Chenin blanc wines (0.3-5.4 mg L<sup>-1</sup>) (Lilly *et al.*, 2000).

Ethyl caprylate is an ester contributed to wine odor (apple-like aroma) (Lilly et al., 2000). It was found in the range of 1.9-3.1 mg L<sup>-1</sup> in Chenin blanc wines (Lilly et al., 2000) and present in lower contents in Monastrell wines, kiwifruit wine, Baga red wine, Mencía wines and sherry wines (Moreno et al., 2005; Rocha et al., 2004; Soufleros et al., 2001; Mateo et al., 2001; Calleja and Falqué, 2005). Ethyl caprylate concentrations of the Sato samples were however not detected in this study.

Ethyl decanoate is produced during the raw material fermentation (Soufleros et al., 2004). It is described as having fruity, grape and brandy to wines (Peinado et al., 2004). From aroma index and GC-O chromatography/olfactometry) analysis, ethyl decanoate impacts odorant of Kalecik Karasé wines (Selli et al., 2006). Ethyl decanoate concentrations of the Sato samples were in the range of 0.66-1.46 mg L<sup>-1</sup>. The results were found at higher concentrations in comparison to Kalecik Karasé wines (0.17-0.31 mg L<sup>-1</sup>) (Selli et al., 2004) whereas its concentration was similar to that of Monastrell wines  $(0.45-0.73 \text{ mg L}^{-1})$  (Mateo et al., 2001).

Diethyl succinate is an ester that is mainly produced from bacterial alterations of various wine components (Soufleros *et al.*, 2001). It is described as having fruity to the wine (Peinado *et al.*, 2006). Diethyl succinate levels of the Sato samples were in the range of 2.88-9.75 mg L<sup>-1</sup> which were higher than those of kiwifruit wines (0.05-1.82 mg L<sup>-1</sup>) (Soufleros *et al.*, 2001), Chenin blanc wines (1.1-1.7 mg L<sup>-1</sup>) (Lilly *et al.*, 2000), Kalecik Karasé wines (ND-0.76 mg L<sup>-1</sup>) (Selli *et al.*, 2004) and wines from grapes (1.1-4.0 mg L<sup>-1</sup>) (Francioli *et al.*, 1999). The results of this study were however similar to those of sherry fino wines (9.42-14.4 mg L<sup>-1</sup>) (Moreno *et al.*, 2005). Low levels of diethyl succinate in the Sato samples suggested that it have no negative effect on the Sato quality.

Ethyl laurate (ethyl dodecylate or ethyl dodecanoate or ethyl laurinate) is described as having sweaty, waxy, floral and soapy to the wine. Ethyl laurate concentrations of the Sato samples were in the range of ND-1.45 mg L<sup>-1</sup>. The compound was found in Portuguese Bagaceiras and Karasé wines at concentrations of 0.85-5.17 and 0.004-0.029 mg L<sup>-1</sup> respectively (Selli *et al.*, 2004, 2006; Silva *et al.*, 1996).

Methanol is not a normal product of alcoholic fermentation. The control of methanol content in alcoholic

beverages is especially important because of its toxicity. Methanol contents in the analyzed samples ranged between 0.96 and 5.15 mg L<sup>-1</sup> which did not exceed the maximum acceptable level (420 mg L<sup>-1</sup>) of fruit wines in Thailand. The methanol concentrations in the Sato were much lower than those found in sherry fino wines (68.6-80.1 mg L<sup>-1</sup>) (Moreno et al., 2005), Mencía wines (121-138 mg L<sup>-1</sup>) (Calleja and Falqué, 2005), kiwifruit wines (485-768 mg L<sup>-1</sup>) (Soufleros et al., 2001), Turkish varietal wines  $(30.5-207.0 \text{ mg L}^{-1})$  (Cabaroglu, 2005), fruit wines and fruit distillate Mouro (371.3-1980.7 mg L<sup>-1</sup>) (Soufleros et al., 2004). The presence of this compound may not influence on sensory of alcoholic drinks as its perception threshold is very high. However, there is some negative effect from a toxicological point of view. Its ingestion in large can produce blindness or even death (Cortés et al., 2005). The high methanol contents are responsible for the splitting of pectic substances to galacturonic acid and methanol (Soufleros et al., 2001; Hernández-Gómez et al., 2003). The low methanol concentrations in all Sato samples indicated that raw materials used for Sato production had low pectin content.

1-Propanol is described as a ripe fruity flavor (Peinado *et al.*, 2006). However, 1-propanol was not detected in the Sato samples but it was found in sherry fino wines (34.8-71.5 mg  $L^{-1}$ ) (Mateo *et al.*, 2001), Chenin blanc wines (17.2-64.1 mg  $L^{-1}$ ) (Lilly *et al.*, 2000) and sparking wines (6.0-10.7 mg  $L^{-1}$ ) (Mamede *et al.*, 2005). The content of 1-propanol in wines is especially influenced by yeast strains responsible for the fermentation (Giudici *et al.*, 1993). Increased 1-propanol concentrations in samples might be the result of a possible microbial spoilage during storage under unfavorable conditions (Apostolopoulou *et al.*, 2005).

1-Butanol is described as a medicinal and phenolic flavor (Peinado *et al.*, 2006). 1-Butanol concentrations of the Sato numbers 1 and 2 were in the range of 0.50-1.29 mg L<sup>-1</sup> which were similar to those of red wines (0.57-0.96 mg L<sup>-1</sup>) (Pérez-Prieto *et al.*, 2003) and Chenin blanc wines (0.3-0.8 mg L<sup>-1</sup>) (Lilly *et al.*, 2000). Higher concentration of 1-butanol was found in the Sato number 3 (10.21 mg L<sup>-1</sup>). In Kalecik Karasé wines, 1-butanol concentration was quite high up to 85.5-108.8 mg L<sup>-1</sup> (Selli *et al.*, 2004). It was reported that 1-butanol was an important higher alcohol in Mouro distillate (Soufleros *et al.*, 2004). However, wines containing high levels of higher alcohols are generally considered of inferior quality.

Isobutyl alcohol (2-methyl-1-propanol) is described as alcohol and solvent flavor (Peinado *et al.*, 2006). Isobutyl alcohol concentrations in alcoholic beverage

quite vary. For the Sato samples, it was detected in the range of 60.01-77.76 mg L<sup>-1</sup>. Similar concentrations were found in Mencía wines (41-64 mg L<sup>-1</sup>) (Calleja and Falqué, 2005) and sherry fino wines (48.0-70.6 mg L<sup>-1</sup>) (Moreno *et al.*, 2005). Higher concentrations were found in sake (110-120 mg L<sup>-1</sup>) (Asano *et al.*, 1999) and Monastrell wines (73-144 mg L<sup>-1</sup>) (Mateo *et al.*, 2001) while lower concentrations were found in Chenin blanc wines (12.5-34.1 mg L<sup>-1</sup>) (Lilly *et al.*, 2000). It is believed that isobutyl alcohol is formed during alcoholic fermentation from the reaction between ethanol and acetic esters (Apostolopoulou *et al.*, 2005).

Isoamyl alcohol (3-methyl-1-butanol) concentrations detected in the analyzed samples were 79.06 to 155.90 mg L<sup>-1</sup>. These were lower than those of sake (196-247 mg L<sup>-1</sup>) (Asano et al., 1999), Monastrell wines  $(246-361 \text{ mg L}^{-1})$  (Mateo et al., 1999) and sherry fino wines  $(300-380 \text{ mg L}^{-1})$  (Moreno et al., 2005). However, the isoamyl alcohol contents in this study were similar to those of Chenin blanc wines (73.2-141.9 mg L<sup>-1</sup>) (Silva et al., 1996) and Kalecik Karasé wines  $(85.50-108.85 \text{ mg L}^{-1})$ . Isoamyl alcohol is contributed to wine aroma and is characterized by fruity attribute (Selli et al., 2004). It is formed during fermentation by deamination and decarboxylation reactions from isoleucine and leucine respectively (Boulton et al., 1996; Kana et al., 1988). Increased concentration of amyl alcohols such as 2-methyl-1-butanol and 3-methyl-1butanol (having an aroma description of alcoholic, sweet and choking) can negatively contribute to the aroma of distillate (Falqué et al., 2001).

4-Penten-1-ol (methylbutan-1-ol) is a higher alcohol which has a positive effect on wine quality in quantities not higher than 500-600 mg  $\rm L^{-1}$ . Higher concentrations of the alcohol cause a negative effect (Soufleros *et al.*, 2001). 4-Penten-1-ol in the Sato samples was only 1.29-58.33 mg  $\rm L^{-1}$  while it was 70-370 mg  $\rm L^{-1}$  in kiwifruit wines (Soufleros *et al.*, 2001). The results suggested that 4-penten-1-ol in the Sato samples had no negative effect on the Sato quality.

4-Methyl-1-pentanol and 3-methyl-1-pentanol were detected in the samples at very low levels (ND-0.01 and 0.06-22.33 mg L<sup>-</sup>,<sup>1</sup> respectively). The higher alcohols were found in Kalecik Karasé wines at concentrations of 15-29 and 50-51 μg L<sup>-1</sup>, respectively (Selli *et al.*, 2004). 3-Methyl-1-pentanol is described as having pungent, solvent and green to wines (Peinado *et al.*, 2006). Low levels of 4-methyl-1-pentanol and 3-methyl-1-pentanol in the Sato samples suggested that they had no negative effect on the Sato quality.

1-Hexanol is described as having herbaceous and wood to wines (Peinado et al., 2006) and it is unfavorable

to wine quality (Selli et al., 2006). 1-Hexanol is not an alcoholic fermentation product (Apostolopoulou et al., 2005) but it is an alcohol originating only from raw material (Soufleros et al., 2001, 2004). It is however considered to be a favorable compound if its concentration is above 5 mg L<sup>-1</sup> but not higher than 100 mg L<sup>-1</sup> (Soufleros et al., 2004). In this study, 1-hexanol concentration of Sato number 2 was 42.28 mg L<sup>-1</sup>. Therefore, the 1-hexanol concentration in this sample is considered to positively affect the flavor of the product. The alcohol was found at higher concentrations in Sato numbers 1 and 3 (217.55 and 106.75 mg L<sup>-1</sup>, respectively). The results obtained from this study were much higher than those of Chenin blanc wines (Lilly et al., 2000), Monastrell wines, Kiwi fruitwines, Karasi wines, Sherry wine and Kalecik Karasé wines (Selli et al., 2004) (0.25-1.63 mg L<sup>-1</sup>). Higher concentrations of 1-hexanol were found in Portuguese Bagaceiras (36-95 mg L<sup>-1</sup>) (Silva et al., 1996). High concentration of 1-hexanol in the Sato numbers 1 and 3 suggested that it caused negative effect on the flavor of the product.

Low concentrations of phenethyl alcohol, which is described as floral, rose-like, sweet, lavender, have positive influence on the wine aroma (Selli et al., 2004; Apostolopoulou et al., 2005). It contributes to the typical flavor characteristics of Mouro fruit to distillate (Soufleros et al., 2004). Phenethyl alcohol concentrations of the Sato samples were in the range of 23.08 to 31.14 mg L<sup>-1</sup>. The results obtained were slightly higher than those of red wines from cv. Kalecik Karasé (12.85-16.20 mg L<sup>-1</sup>) (Selli et al., 2004) and Chenin blanc wines  $(6.0-15.2 \text{ mg L}^{-1})$  (Silva et al., 1996) but lower than those of Baga red wines (65.33 mg L<sup>-1</sup>) (Rocha et al., 2004), red wines (55.08-74.92 mg L<sup>-1</sup>) (Pérez-Prieto *et al.*, 2003), Monastrell wines (75-159 mg L<sup>-1</sup>) (Mateo et al., 2001) and sherry fino wines (52.0-64.1 mg  $L^{-1}$ ) (Moreno et al., 2005).

Caproic acid is described as having rancid, cheese and fatty to the wine (Peinado *et al.*, 2006). Its concentrations of the Sato samples were in the range of ND-3.94 mg  $\rm L^{-1}$ . Similar range of caproic acid concentration was found in Portuguese Bagaceiras, Monastrell wines, Karasi wines and Sherry wine (0.13-4 mg  $\rm L^{-1}$ ) whereas higher concentrations were detected in Chenin blanc wines (5.3-9.1 mg  $\rm L^{-1}$ ).

Isovaleric (3-methylbutanoic acid) and butyric acids are short chain fatty acids which are usually found at low concentrations but the odor is similar in strength string smell to acetic acid (Soufleros *et al.*, 2004). It was noted that they are probably released as intermediate products of long chain fatty acid via yeast metabolism of carbohydrates and can be influenced by insoluble grape solids during fermentation presenting a soapy odor

(Karagiannis and Lanaridis, 2002). However, butyric acid of the Sato samples was not determined in this study. Isovaleric acid was not detected in the Sato numbers 2 and 3 whereas it was detected in the Sato number 1 at concentration of  $5.04 \text{ mg L}^{-1}$ . This concentration was slightly higher than those of kiwifruit wine  $(1.07\text{-}3.94 \text{ mg L}^{-1})$  (Soufleros *et al.*, 2001).

Furfural is formed during heating in acid conditions and/or Maillard reaction due to the dehydration of fermentable sugars (pentoses) (Apostolopoulou *et al.*, 2005). Its odor is reminiscent of bitter almond and cinnamon and it has a toxic character. Furfural was not found in Sato numbers 2 and 3 but it was found in Sato number 1 at concentration of 3.69 mg L<sup>-1</sup>. The results obtained were similar to those of sherry fino wines (ND-4.08 mg L<sup>-1</sup>) (Moreno *et al.*, 2005). Low levels of furfural in this study suggested that furfural have no negative effect on the Sato quality.

It was reported that the higher ratios of esters to alcohols resulted in the production of wines with more fruity character (Mallouchos *et al.*, 2003). Table 1 showed that Sato number 3 had better ratios of esters, acetates and aldehydes to alcohols. The higher ratios of Sato number 3 indicated that this sample had a dominating fruity and floral aroma more than those of Sato numbers 1 and 2, respectively.

One way to quantify odor activity of a compound in an alcoholic beverage is to determine its aroma index (I). The index is calculated by dividing the concentration of the compound in the Sato samples into its perception threshold (Moreno et al., 2005; Cabaroglu et al., 2002). The odor impact of a substance increases in proportion to its aroma index when this value is more than one. Based on these criteria, the above mentioned compounds (particularly those exhibiting the highest aroma index) can be assumed to be those with the strongest odor impact, thereby contributing to a great extent to the aroma of Sato rice wines and reasonably being largely responsible of the sensory profile of these rice wines. Aroma index of the would-be impact odorants of the Sato samples was shown in Table 2.

<u>Table 2: Aroma index (I) of the would-be impact odorants of Sato rice wine</u>

Aroma index (I)

Compounds	Sato 1	Sato 2	Sato 3	Odor description				
Acetaldehyde	0.11	0.28	1.44	Pungent, ripe apple				
Ethyl acetate	21.30	1.28	47.29	Pineapple, varnish, balsamic				
Ethyl butyrate	80.49	163.76	206.81	Strawberry, apple, banana				
Ethyl decanoate	2.15	2.91	1.32	Brandy, fruity, grape				
2-Phenethyl aceta	te 1.93	0.21	0.17	Fruity				
Isobutyl alcohol	1.01	0.80	1.04	Alcohol, solvent				
Isoamyl alcohol	1.32	1.65	2.60	Nail polish, alcohol				
1-Hexanol	197.77	38.44	97.04	Herbaceous, wood				
Isovaleic acid	20.16	ND	ND	Fatty-rancid, cheesy				
Caproic acid	1.31	0.07	ND	Rancid, cheese, fatty				

ND: Not Detectable

#### CONCLUSIONS

The results showed that volatile compositions of the Thai rice wines were comparable to those of the other traditional wines. From the results of all compounds identified in the Sato samples in Thailand using gas chromatograph, 10 compounds were determined to be the most powerful odorants: acetaldehyde, ethyl acetate, ethyl butyrate, ethyl decanoate, 2-phenethyl acetate, isobutyl alcohol, isoamyl alcohol, 1-hexanol, isovaleic acid and caproic acid (Table 2). According to their aroma index and odor descriptor, they seem to be relevant to the aroma of Sato rice wine, originating from both microbial metabolism and raw material.

The proposed methodology, which does not include a sensory test and/or GC-olfactometric (GC-O) studies, seems to be adequate as a preliminary step to establish the would-be impact odorant of the Sato rice wines. However, further steps, such as GC-olfactometric studies are necessary to confirm the impact of the identified odoractive compounds.

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