

## Chapter 5

### Discussion

#### 5.1 Study for the selection of the most optimal method of Krachai-Dam honey wine processing.

##### 5.1.1 Correlation among physical, chemical and pharmaceutical parameters in the first Krachai-Dam honey wine generation (crop year of 2003-2004).

The Spearman's rho correlation showed good correlation between the color values of Krachai-Dam honey wine in L\*a\*b\* system and TP and AOI (Table 5.1). The color value L\* and b\* of wine had high negatively correlated with TP ( $r=-0.900^{**}$  and  $-0.887^{**}$  respectively) and AOI ( $r=-0.879^{**}$  and  $-0.732^{**}$  respectively), whereas color value a\* had positively correlated with TP ( $0.731^*$ ) and AOI ( $0.790^{**}$ ). Moreover, TP value of wine had also positively correlated with AOI value ( $0.837^{**}$ ). Among the color values of wines, high negatively correlation between color value L\* and a\* ( $-0.918^{**}$ ) and high positively correlation between color value L\* and b\* ( $0.859^{**}$ ) were found. These results were in agreement with other reports of red grape wines in the literatures (Campodonico *et al.*, 1998; Fernández-Pachon *et al.*, 2004; Fogliano *et al.*, 1999; Henn and Stehle, 1998; Minussi *et al.*, 2003; Sánchez-Moreno *et al.*, 1999; Sato *et al.*, 1996 and Simonetti *et al.*, 1997). However, there were higher positive correlation between AOI and TP values of red grape wines ( $r^2$  more than 0.900) than Krachai-Dam honey wines ( $r^2=0.701$ ). The positive correlation between AOI and TP values of Krachai-Dam honey wines ( $r^2=0.701$ ) were much lower than red grape wines ( $r^2$  more than 0.900) but were almost the same as white wines ( $r^2=0.7933$ ) (Makris *et al.*, 2003).

**Table 5.1** Correlation coefficients among nine qualitative parameters of Krachai-Dam honey wine in first wine generation (crop year of 2003-2004).

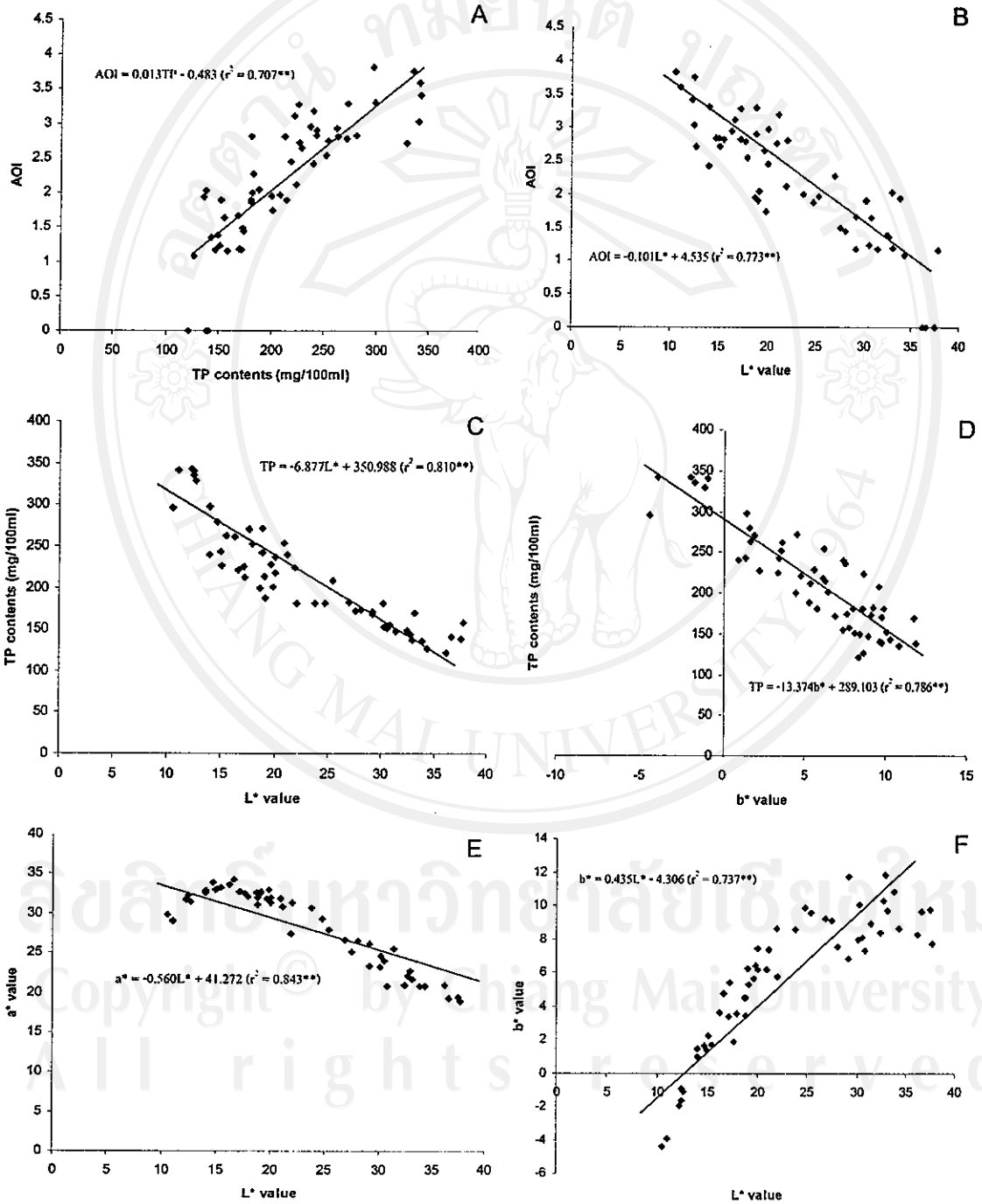
	L*	a*	b*	TSS	ALC	pH	TA	TP	AOI
L*	1.000	-0.918**	0.859**	0.124	0.209	-0.284*	-0.537**	-0.900**	-0.879**
a*		1.000	-0.640**	-0.184	-0.095	0.264	0.484**	0.731**	0.790**
b*			1.000	0.113	0.248	-0.242	-0.449**	-0.887**	-0.732**
TSS				1.000	0.209	-0.183	-0.286*	-0.074	-0.227
ALC					1.000	0.202	-0.508**	-0.350**	-0.164
pH						1.000	-0.193	0.190	0.178
TA							1.000	0.659**	0.623**
TP								1.000	0.837**
AOI									1.000

\*\* Correlation is significant at the 0.01 level.

\* Correlation is significant at the 0.05 level

The linear relationship between AOI and TP, for all Krachai-Dam honey wines (Figure 5.1 (A)), was observed in linear regression equation form of  $AOI=0.013TP-0.483$  ( $r^2=0.701^{**}$ ). It meant that we could use TP for predicting AOI of wines with the coefficient of determination of 70.1%. The other linear regressions were shown in Figure 5.1 (B-F). Their linear regression equations were as followed:

TP	= -6.877L* + 350.988	(r <sup>2</sup> = 0.810**)
AOI	= -0.101L* + 4.535	(r <sup>2</sup> = 0.773**)
a*	= -0.560L* + 41.272	(r <sup>2</sup> = 0.843**)
b*	= 0.435L* - 4.306	(r <sup>2</sup> = 0.737**)
TP	= -13.374b* + 289.103	(r <sup>2</sup> = 0.786**)



**Figure 5.1** The relationships between AOI and TP content (A), AOI and L\* (B), TP content and L\* (C), TP content and b\* (D), a\* and L\* (E) and b\* and L\* (F).

It can be seen that L\* value showed high correlated with TP, AOI, a\* and b\* with the coefficient of determination more than 73.7%, so this L\* value of wines could be used to estimate their TP and AOI. Moreover, the b\* value can also be used for TP estimation with the coefficient of determination of 78.6%. Therefore, this point gave a high benefit from using the L\* and b\* values of wines, which were read easily and quickly by color reader in L\*a\*b\* system, to predict or estimate their TP and AOI, instead of using slow and complicated chemical analytical techniques.

#### 5.1.2 The selection of the most optimal method of Krachai-Dam honey wine processing.

Treatment 163 showed the highest TP (342 mg GAE/100 ml) and AOI (3.42) along with the highest appreciation scores (61.74) from sensory testing. It was the product from 'Rom-Klao' cultivar (dark-purple internal color) with proportion of rhizomes 12.5% w/w; Longan honey and Lalvin V1116 (dry active powder yeast strain) in fermentation process. The appreciation scores of this optimal method were only standard (Margalit, 1996). The explanation in this point was occurred from their strong odor, which was Krachai-Dam characteristics. Therefore, the researcher should improve this weaken-point of these wines.

In addition, the Krachai-Dam honey wine rendered a higher TP and AOI than red wines and much higher than white wines from grapes, which could be described as followed:

The concentration of total phenol as determined by the Folin-Ciocalteu method of the selected combination (number 163) was 342 mg/100 ml GAE or 3,420 mg/l GAE and antioxidant index with  $\beta$ -carotene bleaching (BCB) method of 3.42 or antioxidant activity coefficient of 3,420.

When we considered these values of red and white wines from literatures, it could be found that TP contents of 50 French red wines (Merlot, Cabernet-Sauvignon, Grenache, Syrah and Egiodola varietal wines were varied from 1,847 to 2,600 mg/l GAE (Teissedre and Landrault, 2000); values of 3 monovarietal red wines (Cabernet Sauvignon (2002), Tempranillo (2003) and Syrah (2003) were varied from 1,137 to 2,813 mg/l GAE (Villaño *et al.*, 2006); values of 16 red wines in the south of Spain were varied from 1,378 to 2,360 mg/l GAE (Fernández- Pachón *et al.*, 2004); values of 6 red wines grown in Croatia (Dingac 2001, Babić 2001, Cabernet Sauvignon 1998, Faros barique 1999 and Merlot 1998) were varied from 2,193 to 3,183 mg/l GAE (Katalinic *et al.*, 2004); and values of 6 red wines (Shiraz, Muscat Hamburg (China), Zinfandel, Barbera (2003 and 2004), Muscat Hamburg (2004) and commercial blended red wine (Dong Pa Ya Yen) were varied from 1,498 to 2,432 mg/l GAE (Woraratphoka *et al.*, 2007).

Whereas the TP contents of white (Chardonnay) wine in commercial bottles from Domain Virginie (Béziers, France) was 245 mg/l GAE (Teissedre and Landrault, 2000); value of 17 white wines in the south of Spain varied from 89 to 407 mg/l GAE (Fernández- Pachón *et al.*, 2004); values of 4 white wines (Maraština 2000, Pošip 2001, Traminac 2000 and Graševina 2000) in Croatia varied from 292 to 402 mg/l GAE (Katalinić *et al.*, 2004); values of 26 Greek white wines of 'Appellation of Origin of High quality' varied from 97.6 to 873 mg/l GAE (Makris *et al.*, 2003); and values of 3 white wines (Italia, Chasselar Dore (2003) and Chenin Blanc (2004) from Suranaree University of Technology (SUT) Farm were varied from 306 to 846 mg/l GAE (Woraratphoka *et al.*, 2007). Thus, our selected Krachai-Dam honey wine

rendered higher TP contents than all red wines and much higher values than all white wines referred above.

It was also found that the antioxidant activity coefficient by BCB method of 6 Croatian red wines (Dingac 2001, Babic 2001, Cabernet Sauvignon 1998, Faros 1998, Faros barique 1999 and Merlot 1998) were varied from 2,520 to 2,823, whereas the values of 4 Croatian white wines (Marastina 2000, Posip 2001, Traminac 2000 and Grasevina 2000) varied from 16 to 63 (Katalinic *et al.*, 2004). Thus, our selected Krachai-Dam honey wine was also showed higher antioxidant activity coefficient or antioxidant index than those six red wines and much higher than the 4 white wines referred above.

### 5.1.3 Calculating the estimation of phenolic intake in moderate Krachai-Dam honey wine consumers.

Over the last few years, the consumption of wine in France has fallen considerably. In 1986, the mean consumption was 305 ml/person/day (Darret *et al.*, 1986). This level fell sharply to 180 ml/person/day in 1995 (Boulet *et al.*, 1995) which was used for calculating the estimation of phenolic intake in moderate wine consumers in France in 2000 (Teissedre and Landrault, 2000). Daily intake of total phenolic compound of red wines which comprised of Merlot, Cabernet Sauvignon, Grenache, Syrah and Egiodala were varied from 333 to 468 mg GAE/day/person (based on a daily wine consumption of 180 ml), whereas Chardonnay white wine rendered the daily intake of TP of 44.1 mg GAE/day/person, 10 time less than for red wines. Comparing these data with our Krachai-Dam honey wine, Daily intake of TP of the wine was 615.6 mg GAE/day/person (based on a daily wine consumption of 180 ml), which had approximately 1.5 times higher than red wines and 15 times higher than white wine.

**Table 5.2:** Daily intake of total phenolic compounds from varieties of wines consumption for the French population.

Varieties of wines	Daily intake in mg/day/person
<b>Red wines</b>	
Merlot	360
Cabernet-Sauvignon	468
Grenache	333
Syrah	408
Egiodola	432
<b>White wine</b>	
Chardonnay	44.1

\* Based on a daily wine consumption of 180 ml.

Source: Teissedre and Landrault, 2000.

### 5.1.4 Toxicity of Krachai-Dam honey wines

Chivapat *et al.*, (2004), have studied toxicity of Krachai-Dam. Oral acute toxicity study revealed that LD value of Krachai-Dam powder (KP) was more than 13.33 g/kg in mice and at this dose, no abnormal histopathological changes were found in various visceral organs.

Chronic toxicity study has been performed in six groups of Wistar rats. Group 1 was a water control group and group 2 to 5 were orally given KP at the doses of 20,

200, 1,000 and 2,000 mg/kg/day respectively. Group 6 was a high recovery group, which received 2000 mg/kg of KP, and then KP was withdrawn for 2 weeks. The result revealed that all KP-treated groups have no difference of body weight and health when compared with their control group. Rats receiving 2000 mg/kg KP had some changes; i.e., higher relative weight of liver ( $p < 0.05$ ) which might be due to lower body weight, less eosinophil ( $p < 0.05$ ), higher serum sodium levels than their control groups ( $p < 0.05$ ) but these were still within normal range. In addition female rats receiving the highest dose had higher cholesterol level than the control group ( $p < 0.05$ ). Histopathological examination of visceral organs did not show any abnormality due to KP toxicity.

Daily intake of Krachai-Dam honey wines with no toxicity could be calculated. Dried 'Phurua-10 (Rom-Klao)' Krachai-Dam powder gave 4.82%w/w of ethanolic extract (Sutthanut *et al.*, 2007). Our wines produced from 12.5%w/w of fresh rhizomes or about 4.2%w/w of dried rhizomes, which was equal to 4.2 mg/ 100 ml of wines. Thus, 750 ml/ bottle of wines comprised of 31,500 mg or 31.5 g of dried Krachai-Dam rhizomes.

As for acute toxicity dose, we assume that our body weight was 60 kg, thus  $LD_{50} > 13.3 \text{ g/kg} \times 60 \text{ kg} = 799.8 \text{ g}$ , which equal to consume the amount of 25.37 bottles of wines. However, this value did not include alcoholic toxicity in human.

As for chronic toxicity dose, we assume that our body weight was 60 kg, thus  $> 2,000 \text{ mg/kg/day} \times 60 \text{ kg} = 120 \text{ g/day}$ , which equal to consume the amount of 3.84 bottles of wines/day. We can consume this amount everyday for 6 months. However, this value did not include alcoholic toxicity in human.

Indeed, this research suggested that to drink Krachai-Dam honey wines based on a daily moderate French consumption, which was 180 ml/person/day. In conclusion, the amounts of Krachai-Dam honey wine, which were consumed in this study, had so far from toxicity dose above.

Therefore, these results in part I revealed the new standard formula for honey wine processing, which rendered high TP, AOI, and wine appreciation scores with no toxicity that could be used for promoting the farmers and wine producers. Furthermore, this optimal method could also be used to study the environmental effects, storage methods, and times on wine qualities in part II.

## 5.2 Study on the effects of certain factors on the qualities of Krachai-Dam honey wine production.

### 5.2.1 Correlation among physical, chemical, and pharmaceutical parameters in the second Krachai-Dam honey wine generation (crop year of 2004-2006 and 2005-2006).

The Spearman's rho correlation among the 9 quality parameters of Krachai-Dam honey wine in part II (crop year of 2004-2006 and 2005-2006) showed good correlation between the color value  $L^*$  and  $b^*$  and TP value (Table 5.3). The color value  $L^*$  had slightly high negatively correlated with TP ( $r = -0.635^{**}$ ) whereas color value  $b^*$  had slightly high positively correlated ( $r = 0.607^{**}$ ). However these correlation coefficients were much lower than the first wine generation in part I (crop year of 2003-2004) which were  $-0.900^{**}$  and  $0.731^{**}$  respectively (Table 5.1). Moreover, there was found a slightly high positively correlation between TP and AOI

values ( $r = 0.743^{**}$ ) but its coefficient was much lower than the first wine generation in part I which was 0.837.

There were moderately correlation between the AOI value and color value  $L^*$  ( $r = -0.522^{**}$ ),  $b^*$  ( $r = -0.491^{**}$ ) and  $a^*$  ( $r = 0.551^{**}$ ) and between TP value and  $b^*$  ( $r = -0.482$ ) but these values were also much lower than the coefficients of the wine produced from first wine generation which were  $-0.879^{**}$ ,  $-0.732^{**}$ ,  $0.790^{**}$ , and  $-0.887^{**}$  respectively.

**Table 5.3** Correlation coefficients among nine qualitative parameters of Krachai-Dam honey wines in second wine generation (2004-2006 and 2005-2006).

	$L^*$	$a^*$	$b^*$	TSS	ALC	pH	TA	TP	AOI
$L^*$	1.000	-0.430**	0.611**	0.368**	0.240*	0.457**	-0.342**	-0.684**	-0.563**
$a^*$		1.000	-0.468**	-0.285*	-0.379**	-0.261*	0.253*	0.642**	0.589**
$b^*$			1.000	0.397**	0.335**	0.500**	-0.246*	-0.590**	-0.577**
TSS				1.000	0.402**	0.321**	-0.112	-0.304*	-0.343**
ALC					1.000	-0.058	-0.139	-0.432**	-0.476**
pH						1.000	-0.249*	-0.391**	-0.202
TA							1.000	0.374**	0.333**
TP								1.000	0.743**
AOI									1.000

\*\* Correlation is significant at the 0.01 level.

\* Correlation is significant at the 0.05 level.

Therefore, Krachai-Dam honey wines in the second generation could use only TP of wines for estimating or predicting their AOI. Its linear regression equation was  $AOI = 0.002TP + 0.733$ , ( $r^2=0.522$ ). However, its coefficient of determination was moderate, which was 52.2%. Whereas, each pair of other values gave moderately low coefficient of determination, which were lower than 40.3% ( $r^2=0.403$ ), could not be used to create linear regression equations for estimating their values. Nevertheless, a multiple linear regression equation was also proposed for estimating TP from color value  $L^*$  and  $a^*$ , which was  $TP = -5.487L^* + 4.559a^* + 270.082$ , ( $r^2=0.616$ ).

The Spearman's rho correlation coefficients of Krachai-Dam honey wines, which produced from the second wine generation (2004-2006 and 2005-2006), were much lower than the first one (2003-2004). The distinction between the first wine generation (2003-2004) and the second one (2004-2006 and 2005-2006) were influenced from genotypic and environmental effects. The first wine generation had used the raw materials that were grown in the same plantation area (Phurua area); but varied Krachai-Dam cultivars (Rom-Klao, Nam-Juang and Kheng-Noi-2). Whereas the second one had used the raw materials that were the same cultivar (Rom-Klao); but varied their environmental and practical management, by growing in 4 plantation area, 4 plantation time, 5 harvesting time, 2 crop years, 3 storage methods, and 3 storage times.

The good correlation and linear regression between TP and AOI,  $L^*$ ,  $a^*$  and  $b^*$  values were shown when used raw materials from several Krachai-Dam cultivars but much lower correlation were found instead when the same cultivar was used.

Thus, the environmental effect had more influenced on TP, AOI, and color values of Krachai-Dam honey wines than genotypic effect. Unfortunately, the fluctuation between the results from these two wine generations studied had brought us could not truly use color values ( $L^*$ ,  $a^*$  or  $b^*$ ) for predicting TP or AOI values of

Krachai-Dam honey wines which were produced from raw materials which grown in several plantation areas and used only one cultivar. However, we could use color values ( $L^*$ ,  $a^*$  or  $b^*$ ) to predict only Krachai-Dam honey wines which were produced from raw materials in the same plantation area and several cultivars.

### **5.2.2 The selection of the most optimal method of Krachai-Dam honey wine processing in the second wine generation.**

The wine in treatment 2-28 showed the effects of environmental factors on wine qualities. It was produced from the rhizomes that were grown at 'Maechonluang' area (1,350 m asl) in May, harvested in February and 1-year crop. It gave the highest TP (380 mg GAE/100ml), AOI (1.36) and appreciation scores from sensory testing. Moreover, the cold storage at 13°C, 65% RH within 6 months after harvested could preserve the TP and AOI of wines in all plantation areas.

The TP and AOI values of the second Krachai-Dam honey wine generation, which were produced by using optimal method (combination 163), from raw materials in 1- and 2-year crop (2004-2006 and 2005-2006) were investigated. It was found that the TP values were varied from 213.72 to 389.50 mg GAE/100 ml, whereas the AOI values were varied from 1.08 to 1.44. When comparing these values between the two wine generations which were produced from the 2 crop years, it could be found that the TP values were almost similar with these 2 wine generations but AOI values in second wine generation were much lower than the first one. It might be described that the phenolic compounds, which were the antioxidants in wines that were produced in the first generation, were much higher than in the second one although the total phenolic compounds were almost the same. This study did not analyze each phenolic compound by using GC-MS or HPLC-MS, but analyzed only the total antioxidant activity by BCB method. Indeed, the phenolic compounds of Krachai-Dam honey wines, which were used for health promotion, showed not only their antioxidant activities, but also showed their other pharmaceutical activities, such as anti-stress, adaptogenic, vasodilation and aphrodisiac activities, etc. Therefore, we should continue to study that the other phenolic compounds, which were not antioxidants, whether it might show the other activities mentioned above.

## **5.3 Factors that influenced Krachai-Dam rhizomes used as raw materials for Krachai-Dam.**

### **5.3.1 Correlation among qualitative yields parameters of raw materials that were used to produce the second Krachai-Dam honey wine generation (crop year of 2004-2006 and 2005-2006).**

The Spearman's rho correlation coefficient among the color values  $L^*$ ,  $a^*$  and  $b^*$  and TP and AOI of Krachai-Dam rhizomes, which were used as raw materials in second crop season (Table 5.4), gave high positively correlation between AOI and TP ( $r = 0.884^{**}$ ) and moderately high negatively correlation between color value  $L^*$  and TP and AOI ( $r = -0.692^{**}$  and  $-0.707^{**}$  respectively). However, there were poorly correlation between the color values  $L^*$  and  $b^*$  ( $r = 0.284^*$ ),  $L^*$  and  $a^*$  ( $r = -0.245^*$ ), whereas in the former work, there were found the high correlation between the color value  $L^*$  and  $a^*$  ( $-0.825^{**}$ ),  $L^*$  and  $b^*$  ( $-0.916^{**}$ ) and  $a^*$  and  $b^*$  ( $0.965^{**}$ ) (Pojanagaroon and Kaewrak, 2003). It could be explained that in this second crop season, all treatment combinations were used the same cultivar which was 'Phurua-10 (Rom-klao)' but varied their growing environment and storage practical methods,

whereas in the former work, the 12 cultivars from distinct commercial plantation areas were used to compare. However, we could propose the linear regression for predicting AOI from TP of raw materials, which was  $AOI = 0.012TP + 0.693$ , ( $r^2=0.781$ ); and predicting TP from color value  $L^*$  of raw materials, which was  $TP = -2.386L^* + 143.628$ , ( $r^2=0.479$ ). Nevertheless, a multiple linear regression equation was also proposed for estimating AOI of raw materials from TP and color value  $L^*$  of internal rhizomes, which was  $AOI = 0.010TP - 0.009L^* + 1.107$ , ( $r^2=0.799$ ). This equation had high coefficient of determination of 79.99%.

**Table 5.4** Correlation coefficients among the color values  $L^*$ ,  $a^*$  and  $b^*$ , TP and AOI values of Krachai-Dam rhizomes which were used as raw materials in crop year of 2004-2006 and 2005-2006.

	$L^*$	$a^*$	$b^*$	TP	AOI
L	1.000	-0.245*	0.284*	-0.692**	-0.707**
$a^*$		1.000	-0.163	0.318**	0.200
$b^*$			1.000	-0.503**	-0.502**
TP				1.000	0.884**
AOI					1.000

\* Correlation is significant at the 0.05 level.

\*\* Correlation is significant at the 0.01 level.

### 5.3.2 Correlation among Krachai-Dam honey wine qualities and qualitative yields parameters of their raw materials in the second generation (crop year of 2004-2006 and 2005-2006).

The correlation among qualitative parameters of Krachai-Dam honey wines and their raw materials were studied (Table 5.5). It could be found that there were moderately high positively correlation between color value  $L^*$  of raw materials and  $L^*$  value of wines ( $r = 0.640^{**}$ ), TP of raw materials and TP of wines ( $r = 0.694^{**}$ ) and AOI of raw materials and AOI of wines ( $r = -0.676^{**}$ ). The color value  $L^*$  of raw materials had moderately high negatively correlated with AOI of wines ( $r = -0.701^{**}$ ). Whereas TP of raw materials had positively correlated with AOI of wines ( $r = 0.722^{**}$ ) but had negatively correlated with  $L^*$  value of wines ( $r = -0.637^{**}$ ), and AOI of raw materials had also positively correlated with TP value of wines ( $r = 0.644^{**}$ )

The color value  $L^*$ , TP and AOI of raw materials could be used for estimating TP and AOI of Krachai-Dam honey wines. However, their coefficient of determination of each pair were lower than 50.0%. Therefore, these correlation could give only a trend for predicting TP and AOI of wines from  $L^*$  value of raw materials. We could propose a linear regression between TP of wines and TP of raw materials, which was  $TP_{\text{wine}} = 3.203TP_{\text{raw materials}} + 126.945$ , ( $r^2=0.481$ ). However, its coefficient of determination was moderately low, which was only 48.10%. Nevertheless, a multiple linear regression equation was also proposed for estimating AOI of wines from color value  $L^*$  and TP of raw materials that gave higher coefficient of determination, which was  $TP_{\text{wine}} = 0.005TP_{\text{raw material}} - 0.016L^*_{\text{raw material}} + 1.568$ , ( $r^2=0.616$ ).



**Table 5.5** Correlation coefficients between the color value L\*, a\* and b\*, TP and AOI values of wines and their raw materials.

Krachai-Dam honey wines	Krachai-Dam raw materials				
	L*	a*	b*	TP	AOI
L*	0.640**	0.033	0.165	-0.637**	-0.546**
a*	-0.295*	-0.051	-0.235	0.398**	0.454**
b*	0.568**	-0.101	0.242	-0.576**	-0.547**
TP	-0.556**	0.272*	-0.229	0.694**	0.644**
AOI	-0.700**	0.307*	-0.250	0.722**	0.676**

\* Correlation is significant at the 0.05 level.

\*\* Correlation is significant at the 0.01 level.

### 5.3.3 The qualitative yields of raw materials.

The effects of these factors on qualitative yields of raw materials, which were internal color, TP and AOI of raw materials, were found a good accorded between the wines and their raw materials. However, there were a little distinction between wines and raw materials. The rhizomes, which were kept in cold storage, could retain their TP and AOI values only within 3 months after harvested but significantly reduced in 6 months.

### 5.3.4 The quantitative yields of raw materials.

As for quantitative yields, 'Maechonluang' area gave the significantly highest yields/hectare and weight of rhizomes, which was resulted from long rainy season especially in October and November; higher organic matters, nitrogen and phosphorus mineral contents in soils; and lower maximum, minimum and mean temperatures than other plantation areas. It could be described the meteorological data of 'Maechonluang' area as followed:

1. The altitude of plantation area had influenced on climates of the area. When we considered the meteorological data of the four plantation areas, it could be found that the maximum and minimum temperature of 'Maechonluang' and 'Phurua' areas were almost the same but lower 4-5 °C than 'Phrae' and 'Nakhonphanom' areas throughout the year (Figure 5.2). The maximum/minimum temperature of the last 4-month growing duration (October, November, December, and January) of 'Phurua' and 'Maechonluang' areas were 25.00-32.00/ 9.00-18.00°C.

2. The total monthly rainfall and numbers of rainy day during October to December 2005 were shown in Figure 5.3 and 5.4. 'Maechonluang' area gave much higher rainfall in October (341.7 mm), November (114.8 mm) and December (78.7 mm) than 'Phurua' (74.2, 89.7 and 0.1 mm respectively), 'Phrae' (52.0, 27.1 and 12.03 mm respectively) and 'Nakhonphanom' (18.4, 1.0 and 0.2 respectively). Moreover, 'Maechonluang' area gave much higher numbers of rainy day in October (26 days), November (14 days) and December (6 days) than 'Phurua' (12, 9 and 1 days respectively), 'Phrae' (1, 3 and 1 days respectively) and 'Nakhonphanom' (4, 2 and 1 days respectively).

3. The organic matter contents in soil of 'Maechonluang' area gave much higher organic matter contents (4.18%) than the other areas (1.09-1.93%) and have the highest N and P contents.

4. The types of soil at 'Maechonluang' area were sandy clay loam and sandy clay with lower pH than other areas.

Therefore, the long rainfall duration; lower maximum/minimum temperature; high organic matters, N and P contents in soil; sandy clay loam and sandy clay soil types; and lower soil pH of the plantation area could have high influenced on quantitative yields of 'Maechonluang' area. As for the highest TP and AOI in rhizomes, it could be described as the same.

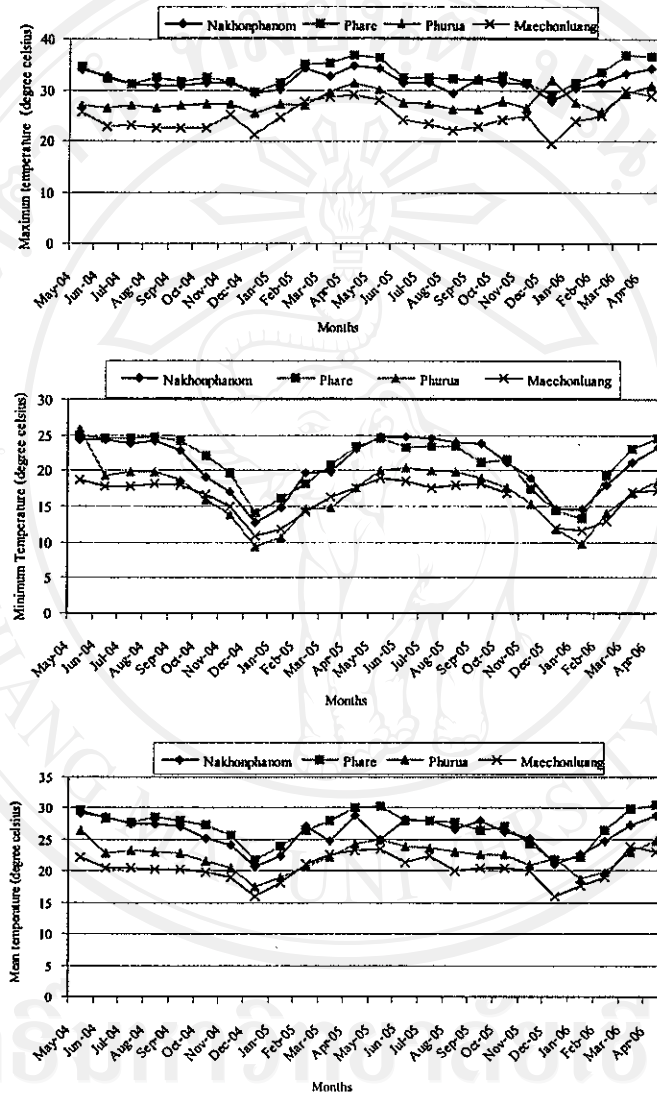


Figure 5.2 Maximum, minimum and mean temperatures during crop season 2004-2006 in four plantation areas.

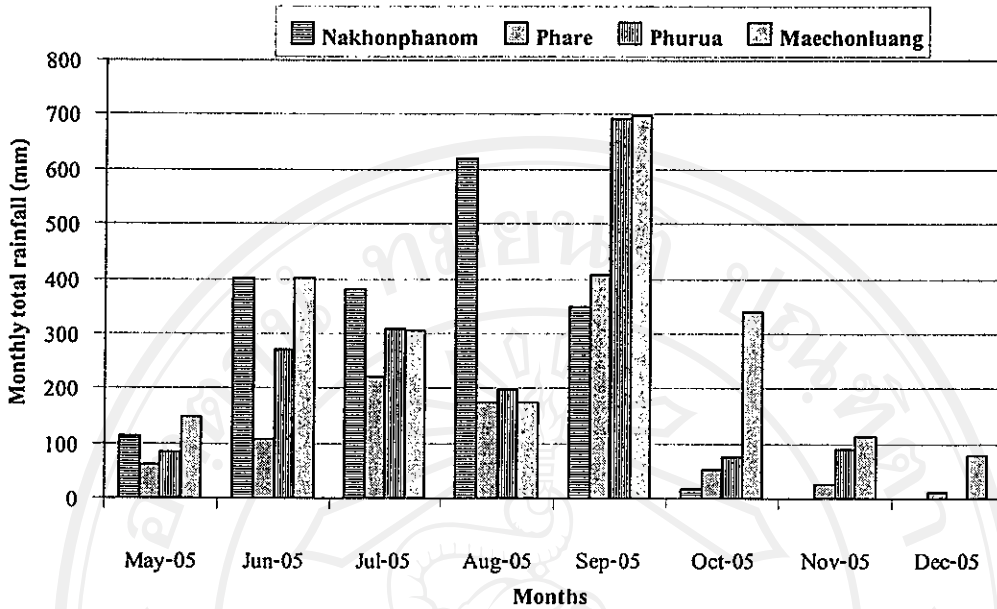


Figure 5.3 monthly total rainfalls during May to December 2005.

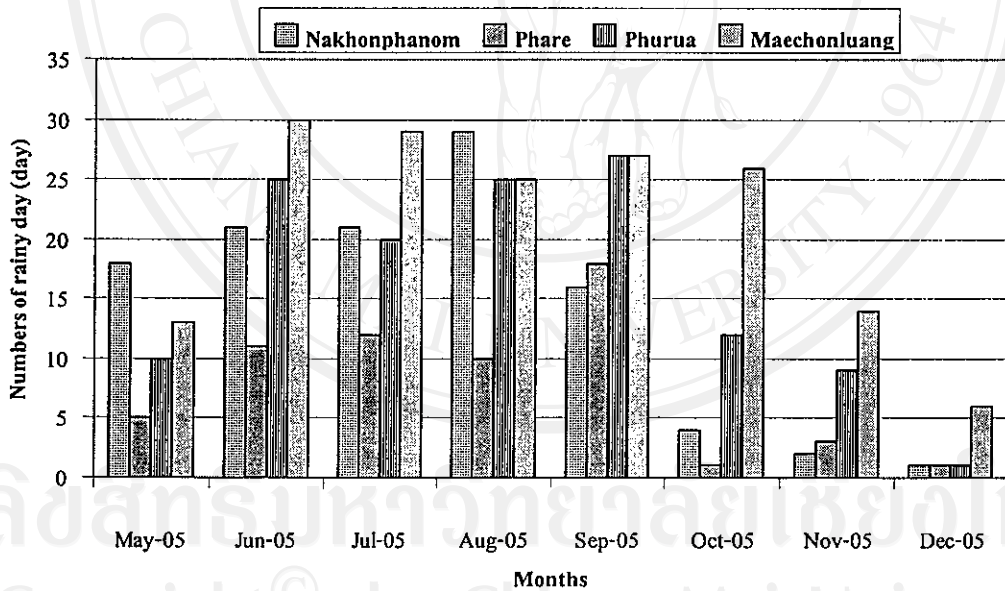


Figure 5.4 Numbers of rainy day during May to December 2005.

The last notification was 2-year crop gave the quality of rhizomes as the same as 1-year crop, but gave significantly higher quantitative yields than 1-year crop. Thus, the farmers must justify between growing expenses from growing 2 times and high risk from *Ralstonia solonacaerum* when growing 2-year crop standing.

#### 5.4 To summarise and to draft the Good Agricultural Practices (GAP) of Krachai-Dam for Krachai-Dam honey wines purposes.

This research revealed the standardization of Krachai-Dam rhizomes as followed: The cultivar that had higher level of internal rhizomes' color gave the higher level of TP and AOI in Krachai-Dam honey wines, which was 'Phurua-10 (Rom-Klao)'. The suitable area was an area with a higher elevation than 950 m asl that had maximum/minimum temperature of 25-30/ 9-18°C. May to June were suitable planting months while January to February were suitable harvesting times. The processing immediately after harvesting or keeping in cold storage for 6 months was also suggested.

Roughly, these researches could be used to draft the GAP of Krachai-Dam for Krachai-Dam honey wines purpose, and could be summarized from this study as followed:

**Scientific name:** *Kaempferia parviflora* Wall. ex Baker.

**Habitat:** Krachai-Dam prefers cold and humid climate, and can be grown in mostly all types of soil, but the best is in sandy clay loam and sandy clay soil with pH of 4.0-5.0. High N and K contents in soils are recommended. Commercial cultivation areas are in uplands of Loei, Phitsanulok, Phetchabun, Nan, Tak, Chiangrai, and Kanchanaburi provinces.

**Krachai-Dam cultivars:** The best cultivar is 'Phurua-10 (Rom-klao)', which has dark purple internal color of rhizomes and gives the highest total phenolic contents and antioxidant index.

**Plantation area:** The suitable area is higher than 200 m asl, but the most suitable area should higher than 900 m asl. The areas should have minimum temperature in a range of 9-18°C, maximum temperature in a range of 25-30°C and have long rainfall duration with hard raining in October and November that influenced on yields. The area should have never been grown zingiberaceons plants or have never been grown within 3 years before planting in order to protect our plants from *Ralstonia solonacaerum*.

**Planting time:** The most suitable planting time is early May, but rhizomes can be grown until late June. However, they cannot be grown in early August; otherwise, their yields will be very low.

**Cultivation:** Propagation can be used by the rhizome cuttings technique, and then placing them in a seedbed with sand. They can be grown on any soils but thrive best on sandy clay loam and sandy clay soils. Land should be prepared and left for a period of about two weeks and then a raised bed of 1.5 m wide should be made. In the field, the rhizomes, with at least two buds, are usually planted, 10-15 cm deep on raised bed, 40-50 cm between rows with 20-30 cm spacing between plants.

**Maintenance:** Water should be provided after planting until sprouts appear. and in early winter (October to November) in an arid zone. Hay covering over the ground to conserve moisture is also suggested. Fertilizers (15-15-15) are applied to newly plants, 2 and 3 months after growing. Hand weeding should be made one month after planting and every month thereafter. Finally, high potassium fertilizers (13-13-21) are applied to mature plants in September for promoting rhizomes growth and their secondary metabolites accumulation. There is no need to spray any pesticide as 'Krachai-Dam' does not have any serious pest. The most dangerous plant pathology of them is *Ralstonia solonacaerum* that have no remedy to eradicate.

**Harvesting:** The rhizomes can be collected by digging with a hoe at 8-9 months after planting, when all leaves turn yellowish-brown and absolutely dry. Cut off the dry leaves and roots, and then wash off adhering soils.

**Post-harvest Handling:** Washing the rhizome thoroughly, and then dry by spreading on the ground or keeping in net bags in a shaded and air-ventilated place. The long-term storage more than 6 months, the keeping in cold storage at 13 C, 65% relative humidity is needed for maintaining their chemical and pharmaceutical qualities and reducing weight loss.

**Quality of Krachai-Dam rhizomes as raw materials:** Good Krachai-Dam rhizomes should have  $L^* = 33.03-38.61$ ,  $a^* = 5.93-13.59$ ,  $b^* = -1.15-5.74$ ,  $TP = 41.93-69.38$  mg/ml of ethanolic extract,  $AOI = 1.25-1.51$ , weight of rhizomes = 21.39-44.23 grams, production per hectare = 1,792.63-4,740.75 kilograms (286.82-758.52 kilograms/rai)

**Quality of Krachai-Dam honey wines:** Good Krachai-Dam honey wines should have  $L^* = 9.80-16.91$ ,  $a^* = 26.83-34.20$ ,  $b^* = -8.98-5.56$ ,  $TP = 293.50-389.50$  mg/100 ml GAE and  $AOI = 1.26-3.42$ .

### 5.5 Further studies

1. The most important reasons for consuming Krachai-Dam product were aphrodisiac uses (as 'Sildenafil (Viagra)'), and adaptogenic and anti-stress uses (as 'ginseng'). This study produced Krachai-Dam honey wine and tested only 'total phenolic compound contents' and 'antioxidant activity of wine but did not study these two necessary activities above. Therefore, further study should analyzes the chemical components which shown aphrodisiac and anti-stress activities in wines. The correlation among  $L^*$ ,  $a^*$ ,  $b^*$ ,  $TP$  and  $AOI$  with the two necessary activities above should also be studied. The results of this point could be used for consumer purchasing decision upon this product.

2. The  $AOI$  of the second wine generation had much lower than the first generation. Thus, further study should analyze each chemical component in wine, which give antioxidant activity by GC-MS techniques. These details reveal that which chemical components could be changed followed the seasonal changes.

3. The distinction between the correlation of  $L^*$ ,  $a^*$ ,  $b^*$ ,  $AOI$  and  $TP^*$  in the first and the second wine generation that might be occurred from genetically and environmental effects. The further study should be made by using three Krachai-Dam cultivars, which will be grown in several plantation areas. More data will be accurate in order to predict equation for  $TP$  and  $AOI$  values of Krachai-Dam honey wines.

4. This study did not make on the storage time of Krachai-Dam honey wine. Thus, further study should be made the suitable storage time that can sustain  $AOI$  and  $TP$  values of wines. Moreover, the chemical components that give aphrodisiac and anti-stress activities should also be studied on their suitable storage time.

5. Roughly, Good Agricultural Practice of Krachai-Dam in this study was drafted from some important factors such as cultivars, plantation area, planting month, harvesting time etc. Thus, further study should be made on other factors such as pests and diseases management, soil fertilities, soil management, fertilizers and irrigation uses etc. After that, the complete GAP drafts must be tested in the farmer fields before pronounced by Department of Agriculture (DOA).