CHAPTER 3
RESULTS

3.1 Wine aging process

Prepared wood chips after cut, sliced and roasted are shown in figure 1. It tokes about 30 minutes for roasting without burning. The roasted woods were submitted for storage in sealed glass containers to prevent oxidation by oxygen and moisture.

(1a) (1b) (1c)

Figure 3.1 Wood chips, (1a) wood timber, (1b) sliced wood, (1c) roasted wood.

Six kinds of roasted wood chips were added into fresh wine (from Maechan Winery) with the ratio of 5g : 1 l wine in 750 ml wine bottles. After aging for 120 days, wine samples were transferred to plastic bottles. The wines obtained were subsequently tasted by trained examiners. The aged wine are shown in figure 3.2.

(a) (b)

Figure 3.2 Wine samples (a) red fresh wine from Maechan Winery (b) red wind after aging with roasted wood chips.

3.2 Wine Sensory Tasting

The scores of several kind ofwine tasted by 50 volunteers is shown in Table 3.1.

Table 3.1 Average satisfactory score by 50 volunteers

Aged wine type	Longan aging	Luna nut aging	Drumstick aging	Neem aging	Black poum aging	Oak aging
Average Score	18.50±0.46	14.95±0.76	17.73±0.44	15.20±0.32	14.62±0.30	17.63±0.38

* Score detail in the appendix.

The results showed that aged wine using Longan wood chip has the highest score of 18.50. Drumstick and Oak aged wines also give good results with the score of 17.73 and 17.63 respectively. In conclusion, wine aged with Longan and Drumstick wood chips were expected to have agreeable acceptance compatible to wine aged with oak wood chip.

From the result, the ANOVA and SPSS computer program was applied to evaluate the score difference of each wine samples. Results of one-way ANOVA analysis showed in table 3.2

Table 3.2 One-way ANOVA analysis of wine samples evaluation score.

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	720.159	5	144.032	655.816	.000
Within Groups	64.569	294	.220		
Total	784.727	299			

From the table, sig. value was .000 which 0.05, indicate that there is at least one wine sample that has the different score compared to other samples. After that, one-way t-test was used to analyze the contrast of data score compare between two types of sample. The result of pair t-test showed in table 3.3

Table 3.3 One-way variance analysis between each pair data group with t-test.

Contrast Between each data group		**Value of Contrast**	**Std. Error**	**t**	**P**
1st Wine sample	2nd Wine sample				
Longan	Luna nut	3.553	0.126	28.235	0.000 +
	Drum stick	0.773	0.090	8.610	0.000 +
	Neem	3.300	0.079	41.942	0.000 +
	Black poum	3.880	0.078	50.049	0.000 +
	Oak	0.867	0.084	10.337	0.000 +
Luna nut	Drum stick	-2.780	0.125	-22.319	0.000 -
	Neem	-0.253	0.117	-2.169	0.034 -
	Black poum	0.327	0.116	2.816	0.006 +
	Oak	-2.687	0.120	-22.329	0.000 -
Drum stick	Neem	2.527	0.077	32.985	0.000 +
	Black poum	3.107	0.075	41.196	0.000 +
	Oak	0.093	0.082	1.140	0.257
Neem	Black poum	0.580	0.062	9.396	0.000 +
	Oak	-2.433	0.069	-35.013	0.000 -
Balck poum	Oak	-3.013	0.068	-44.192	0.000 -

*Sign + behind P-value indicate that the first wine sample score was higher than second score, the sign – behind P-value indicate that the first wine sample score was lower than second score, if no sign was add behind P-value indicate that first and second wine sample scores were not significantly different not difference (P-value more than 0.05)

The results of t-test in table 3.3 that confirm the evaluation score of Longan wood aged wine, present the highest score when compared with all other wine samples because P-value form data analysis versus other samples were 0.00+.

3.3 Volatile compounds analysis

The volatile chemical compositions analysis by SPME-GC-MS method for the six types of aged wine were shown in Figure 3.3 to 3.8.

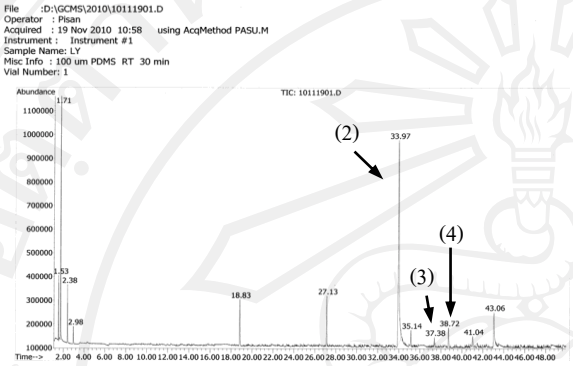

Figure 3.3 TIC chromatogram of Longan wood aged wine.

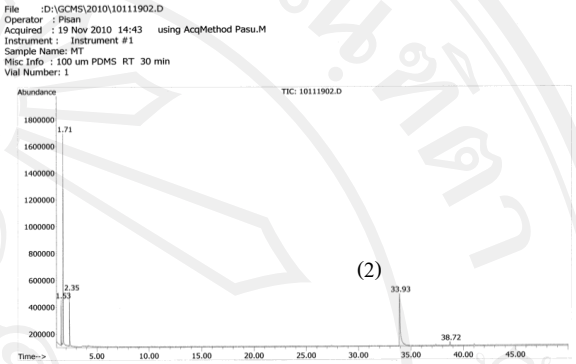

Figure 3.4 TIC chromatogram of Luna nut wood aged wine.

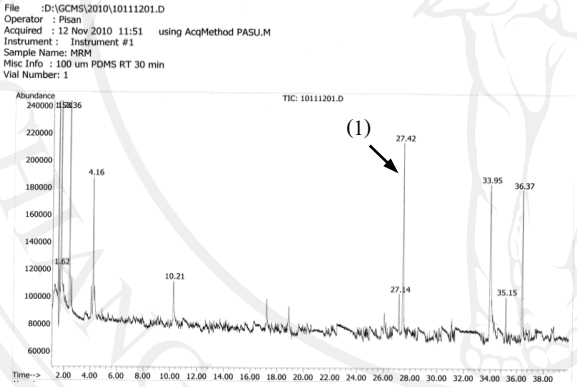

Figure 3.5 TIC chromatogram of Drumstick wood aged wine.

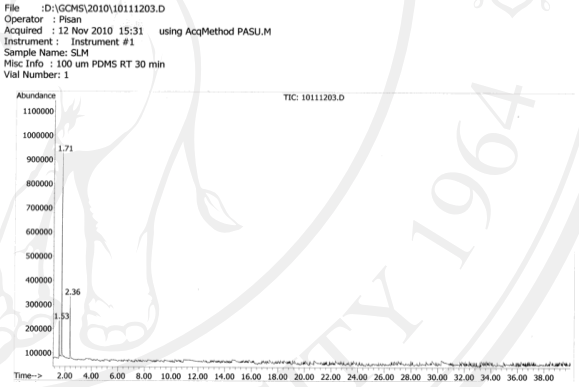

Figure 3.6 TIC chromatogram of Neem wood aged wine.

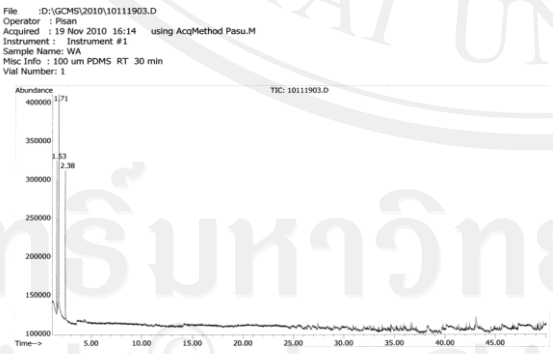

Figure 3.7 TIC chromatogram of Back poum wood aged wine.

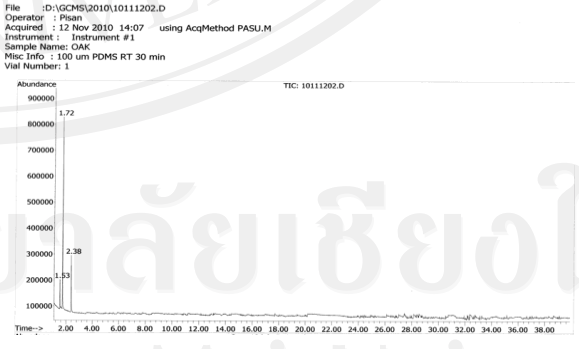

Figure 3.8 TIC chromatogram of Oak wood aged wine.

All identification compounds were shown in table 3.4

Volatile compounds of interest in six types of wine are listed in Table 3.4.

Table 3.4 List of the interested volatile compounds found in wine samples.

Peak No.	RT, min	Compounds	Percent relative composition					
			Longan	Luna nut	Drumstick	Neem	Black poum	Oak
(1)	27.42	Ethyl octanoate	-	-	4.5	-	-	-
(2)	33.97	Eugenol	45.84	44.57	4.23	-	-	-
(3)	37.38	β-caryophyllene	1.22	-	-	-	-	-
(4)	38.72	α-humulene	3.02	1.98	-	-	-	-

Follow the table 3.4, eugenol, β-caryophyllene and α-humulene were found in Longan wood aged wine. In Luna nut wood aged wine, the interested compounds were eugenol and α-humulene. The compounds that found in Drumstick wood aged wine were ethyl octanoate and eugenol. But in case of Neem wood aged wine, Black poum wood aged wine and Oak wood aged wine, we could find that the interested compounds were not detected even though expanded scale of the chromatogram was done. From the results, α-humulene and β-caryophyllene, which were naturally ocurring sesquiterpenes found in the hop essential oil, could be detected in Longan wood aged wine. Hop essential oil was important in beer production to give wood flavor. Eugenol, which was a product formed by decomposition of wood, was also presented. Ethyl octanoate was the fermentation product in wine and was described as fruit flavor.

3.4 Antioxidant activity of wine samples

Antioxidant activity of each wine sample with DPPH method was carried out by preparation at various concentrations (0.5 – 9 % v/v) of wine samples. The reaction was done by adding DPPH reagent and determind the absorbance at 517 nm. The results were shown in table 3.5–3.10.

Table 3.5 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Longan wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\bar{X}$	SD
0	0.2037	0.2037	0.2037	100.00	100.00	100.00	100.00	-
0.5	0.0552	0.0557	0.0555	72.90	72.66	72.75	72.77	0.12
1	0.0967	0.0969	0.0961	52.53	52.43	52.82	52.59	0.20
3	0.1136	0.1136	0.1137	44.23	44.23	44.18	44.21	0.03
5	0.1287	0.1294	0.1291	36.82	36.48	36.62	36.64	0.17
7	0.1433	0.1434	0.1433	29.65	29.60	29.65	29.63	0.03
9	0.1619	0.1621	0.1620	20.52	20.42	20.47	20.47	0.05

Table 3.5, IC_{50} value could be determined by plotting sample concentrations versus absorbance values. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.9.

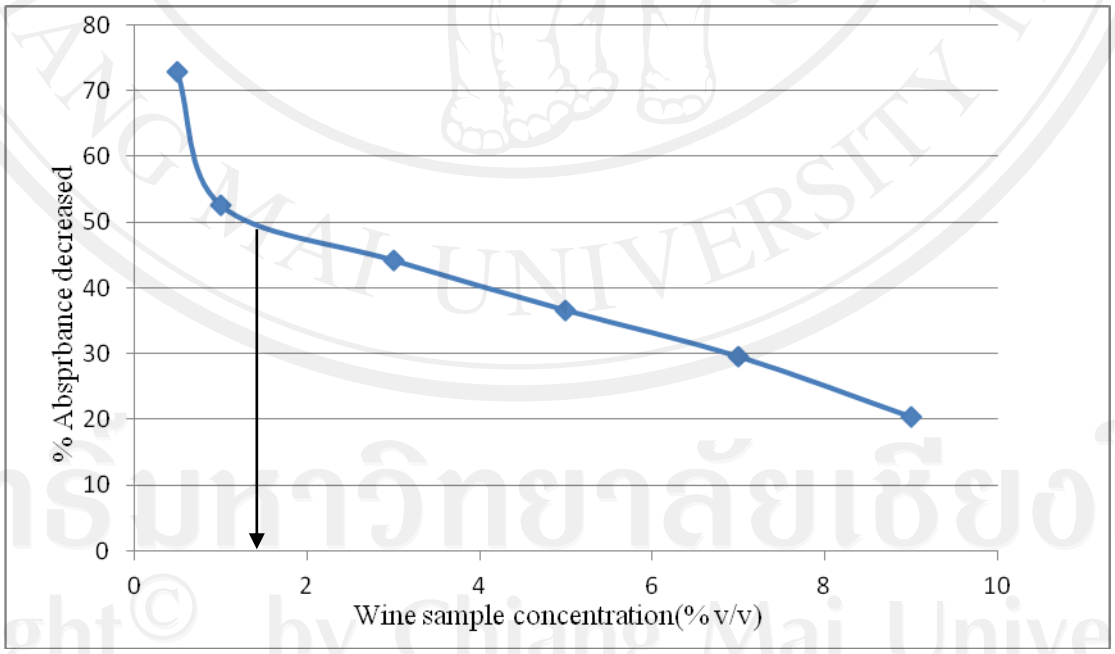

Figure 3.9 The relationship plot of wine concentrations versus absorbance at 517 nm of Longan wood aged wine. The concentration at the decrease of absorbance value to 50% is 1.62% v/v.

Table 3.6 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Luna nut wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\bar{X}$	SD
0	0.2867	0.2867	0.2867	100.00	100.00	100.00	100.00	-
0.5	0.0503	0.0502	0.0504	82.46	82.49	82.42	82.46	0.04
1	0.0631	0.0607	0.0625	77.99	78.83	78.2	78.34	0.44
3	0.0752	0.0702	0.0732	73.77	75.51	74.47	74.58	0.88
5	0.1115	0.1163	0.1144	61.11	59.43	60.10	60.210	0.85
7	0.1433	0.1431	0.1429	50.19	50.09	50.16	50.15	0.05
9	0.1648	0.1659	0.1659	42.52	42.13	42.13	42.26	0.23

Table, 3.6 IC_{50} value could be determined by plotting sample concentrations versus absorbance values. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.10.

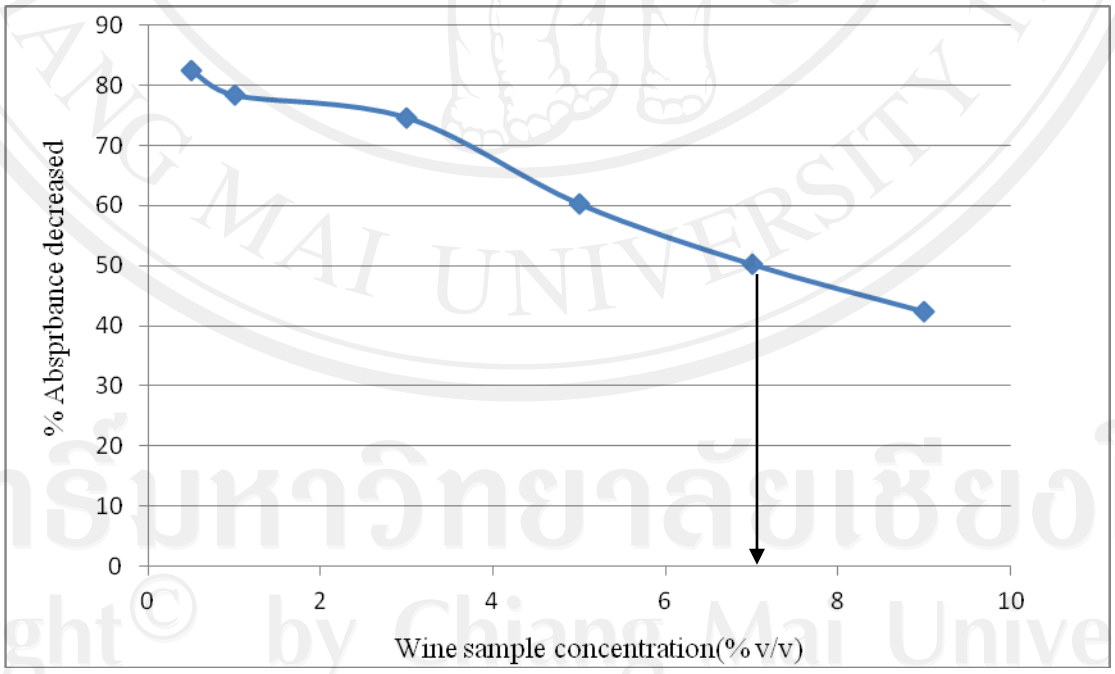

Figure 3.10 The relationship plot of wine concentrations versus absorbance at 517 nm of Luna nut wood aged wine. The concentration at the decrease of absorbance value to 50% is 7.20% v/v.

Table 3.7 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Drumstick wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\bar{X}$	SD
0	0.2850	0.2850	0.2850	100.00	100.00	100.00	100.00	-
0.5	0.0444	0.0487	0.0451	84.42	82.91	84.18	83.84	0.81
1	0.0726	0.0702	0.0704	74.53	75.37	75.3	75.07	0.47
3	0.0801	0.0852	0.0841	71.89	70.11	70.49	70.49	0.94
5	0.1002	0.1071	0.1035	64.84	62.42	63.68	63.65	1.21
7	0.1435	0.1431	0.1433	49.65	49.79	49.72	49.72	0.07
9	0.1725	0.1775	0.1742	39.47	37.72	38.88	38.69	0.89

Table 3.7, IC_{50} value could be determined by plotting sample concentrations versus absorbance value. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.11.

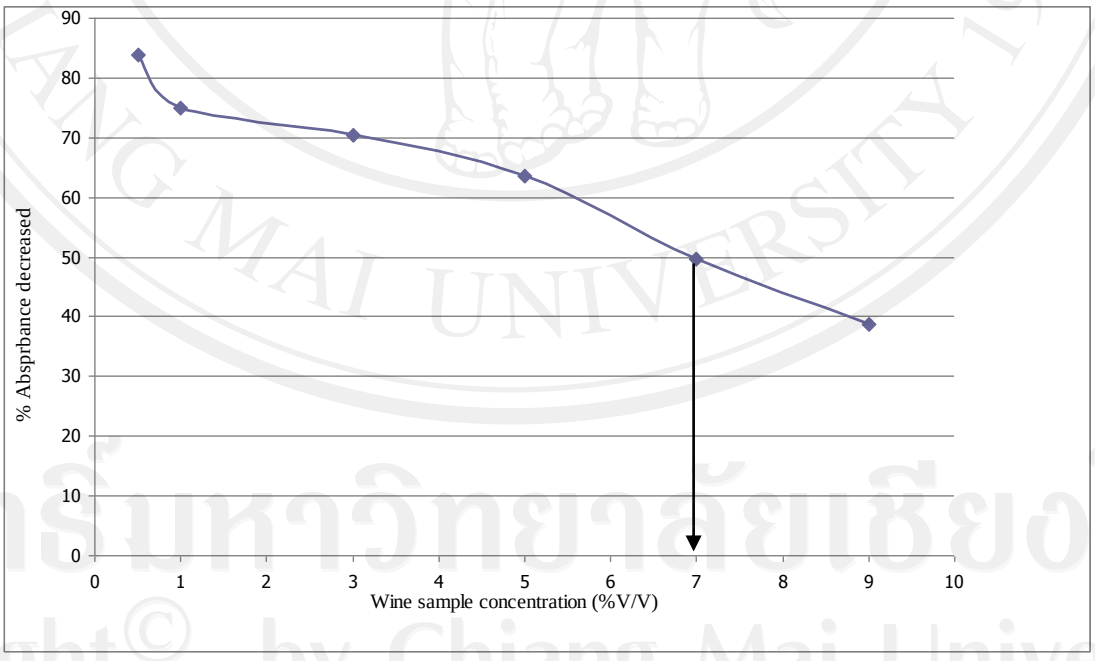

Figure 3.11 The relationship plot of wine concentrations versus absorbance at 517 nm of Drumstick wood aged wine. The concentration at the decrease of absorbance value to 50% is 7.11% v/v.

Table 3.8 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Neem wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\overline{X}$	SD
0	0.2029	0.2029	0.2029	100.00	100.00	100.00	100.00	-
0.5	0.0430	0.0433	0.0430	78.81	78.66	78.81	78.76	0.09
1	0.0632	0.0630	0.0630	68.85	68.95	68.95	68.92	0.06
3	0.0715	0.0716	0.0715	64.76	64.71	64.76	64.74	0.03
5	0.0965	0.0966	0.0965	52.44	52.39	52.44	52.42	0.03
7	0.1213	0.1216	0.1214	40.22	40.07	40.17	40.15	0.08
9	0.1432	0.1435	0.1430	29.42	29.28	29.52	29.41	0.12

Table 3.8, IC_{50} value could be determined by plotting sample concentrations versus absorbance value. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.12.

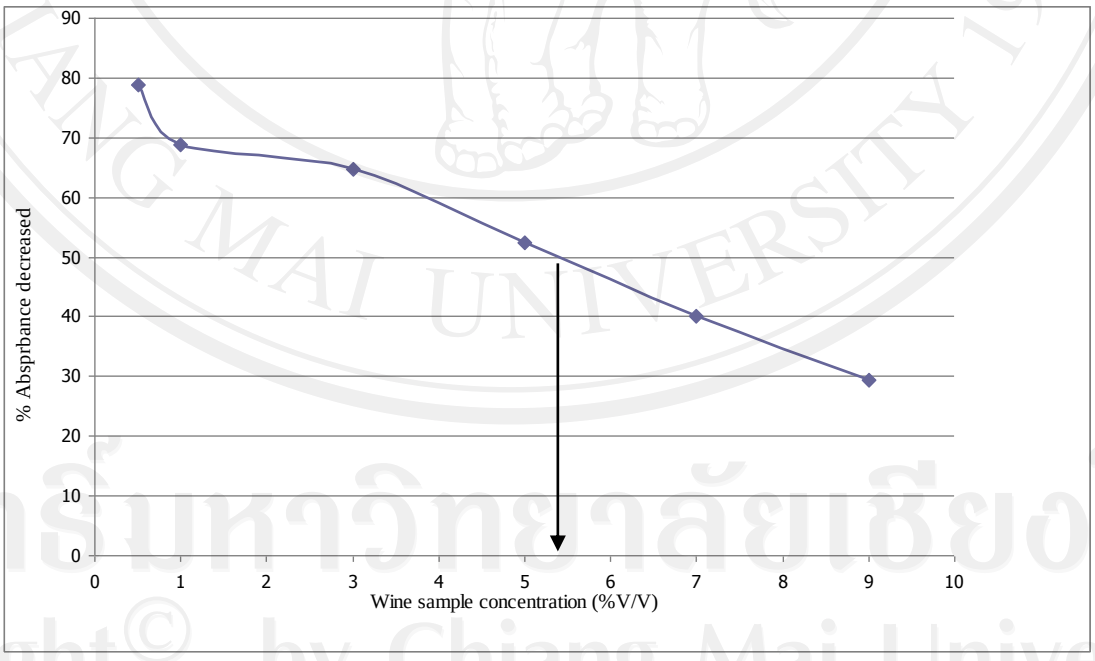

Figure 3.12 The relationship plot of wine concentrations versus absorbance at 517 nm of Neem wood aged wine. The concentration at the decrease of absorbance value to 50% is 5.40%v/v.

Table 3.9 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Black poum wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\bar{X}$	SD
0	0.2820	0.2820	0.2820	100.00	100.00	100.00	100.00	-
0.5	0.0549	0.0588	0.0532	80.53	79.15	81.13	80.27	1.02
1	0.0602	0.0608	0.0607	78.65	78.44	78.48	78.52	0.11
3	0.0652	0.0658	0.0651	76.88	76.67	76.91	76.82	0.13
5	0.1088	0.1089	0.1090	61.42	61.38	61.35	61.38	0.04
7	0.1472	0.1438	0.1436	47.8	49.61	49.08	48.83	0.93
9	0.1688	0.1619	0.1625	40.14	42.59	42.38	41.70	1.36

Table 3.9, IC_{50} value could be determined by plotting sample concentrations versus absorbance value. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.13.

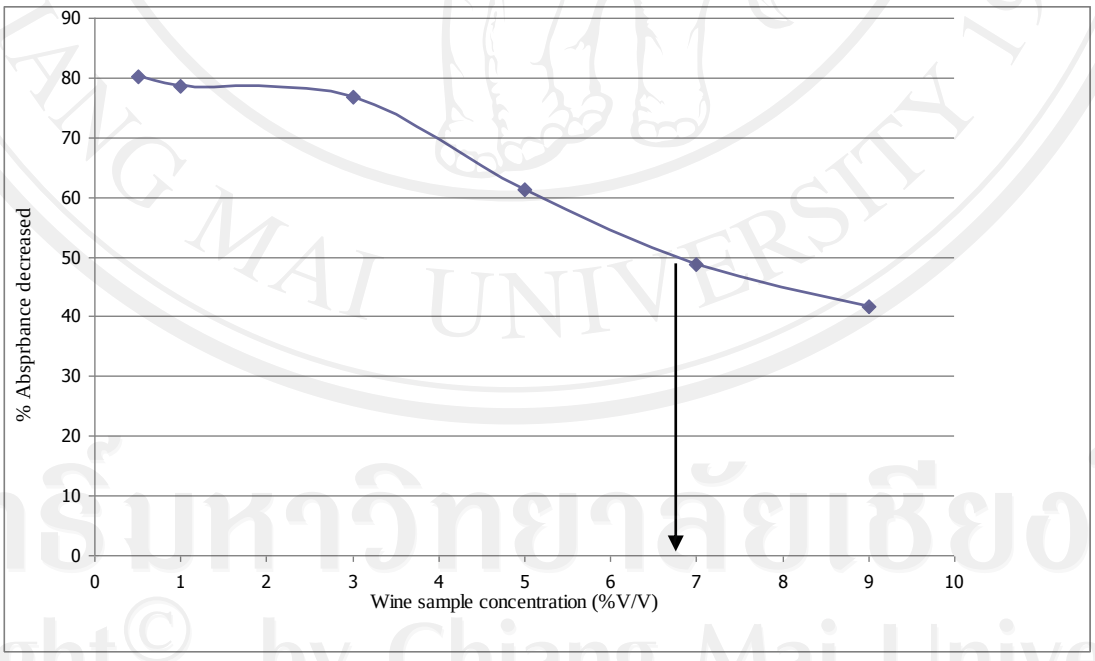

Figure 3.13 The relationship plot of wine concentrations versus absorbance at 517 nm of Black poum wood aged wine. The concentration at the decrease of absorbance value to 50% is 6.76 %v/v.

Table 3.10 Absorbance at 517 nm and percentage of absorbance decrease in antioxidant activity test of Oak wood aged wine.

Wine sample conc. (%v/v)	A_{517}			Percentage of the absorbance decreases.				
	1	2	3	1	2	3	$\bar{X}$	SD
0	0.2831	0.2831	0.2831	100.00	100.00	100.00	100.00	-
0.5	0.0511	0.0512	0.0509	81.95	81.91	82.02	81.96	0.06
1	0.0621	0.0619	0.0620	78.06	78.13	78.10	78.10	0.04
3	0.0732	0.0724	0.0727	74.14	74.43	74.32	74.30	0.15
5	0.1029	0.1014	0.1056	63.65	64.18	62.70	63.51	0.75
7	0.1238	0.1231	0.1235	56.27	56.52	56.38	56.39	0.13
9	0.1489	0.1487	0.1473	47.40	47.40	47.97	47.59	0.33

Table 3.10, IC_{50} value could be determined by plotting sample concentrations versus absorbance value. The point of the concentration able to decrease the absorbance value to 50% is IC_{50}. The plotted graph for the determination of IC_{50} was shown in figure 3.14.

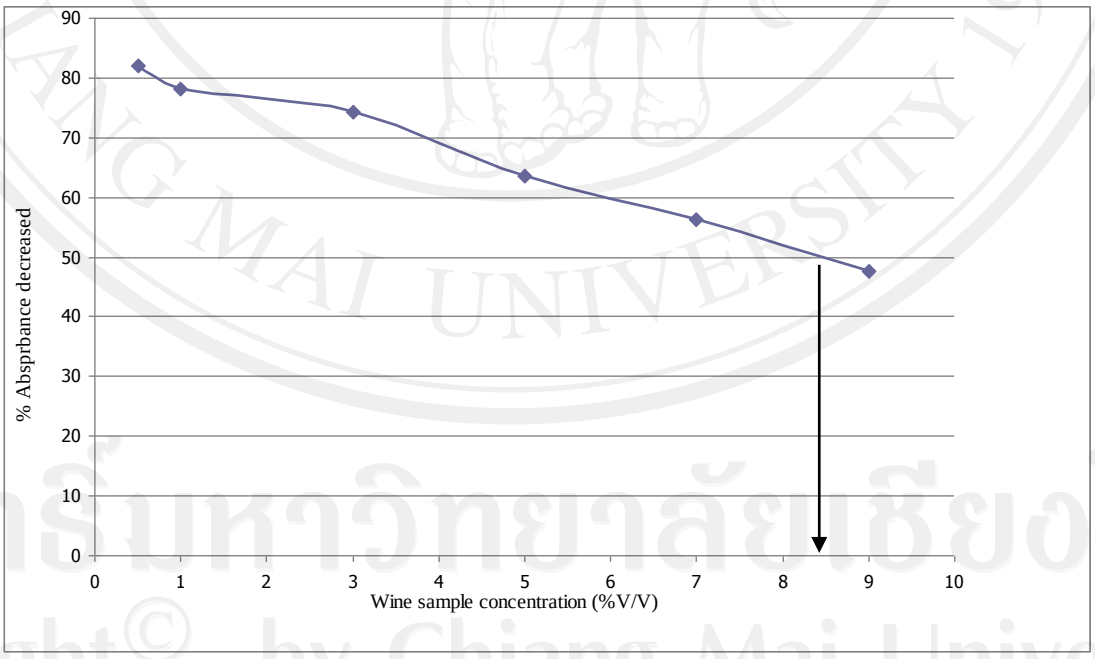

Figure 3.14 The relationship plot of wine concentrations versus absorbance at 517 nm of Oak wood aged wine. The concentration at the decrease of absorbance value to 50% is 8.47 %v/v.

The results from figure 3.9 – 3.14 can summarize the IC_{50} of six wine samples as in the table 3.11.

Table 3.11 The results of IC_{50} of six kinds of wine samples

Sample	IC_{50} (%v/v)
Longan wood aged wine	1.62±0.10
Luna nut wood aged wine	7.20±0.42
Drumstick wood aged wine	7.11±0.73
Neem wood aged wine	5.40±0.07
Black poum wood aged wine	6.76±0.60
Oak wood aged wine	8.47±0.24

Note: low IC_{50} is related to high antioxidant activity

The aged wine sample which has a low IC_{50} value indicate that its antioxidant activity is high. Conversely, if the IC_{50} is high the wine sample has a low antioxidant activity. From the table 3.9, Longan wood aged wine shows the IC_{50} of 1.62 %v/v. Therefore this wine sample has the highest antioxidant activity comparing to other types of wine samples. This result was wimilar to the previous research which found that β-caryophyllene was responsible for the major antioxidant antioxidant activity.