



## RESEARCH ARTICLE

## Potentiality of Using Teak (*Tectona Grandis*) Leaf to produce Antioxidant Herbal Tea

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**ARTICLE INFO****ABSTRACT**

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This research aimed to develop a healthy herbal tea from teak leaf in order to employ non-timber part of teak wood to produce high value products. From previous investigation elsewhere, the reports confirmed that teak leaves have benefits from its active ingredients and the medicinal properties are clearly written. However, the knowledge and processing technology of making teak leaf herbal tea is still limited. Researchers decided to opt the existing commercial tea process with slightly modifications and two prototype of herbal tea, i.e. green tea and oolong tea, were produced. The yield is highly variable depending on the process and the humidity of the product with 22.55- 87.5 0%. The free radical scavenging activity of the teak tea using DPPH method was 3,398.94-6,927.25 µg vitamin c equivalent where the inhibition rate was 39.04-77.74%. This implied that teak leaf has potentiality to produce herbal tea with developed process and its antioxidant effect still remained in the product.

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**INTRODUCTION**

Teak is one of the most high value hardwood and grow in various tropical region, for example, Asia, Africa, Latin America and Oceania and its wood has been use for construction and furniture building (Ball et al., 1999). Thai teak is an extremely good quality that accepted as one of the best teak in the world, popular in the market, has a high price and great economic potential. For last two decade, the natural teak wood was dwindled significantly and many turn their interest to establish commercial teak plantation (Pandey and Brown, 2000). Although the government has a policy to promote teak planting continuously throughout the year, the teak business has stagnated due to many factors i.e. inflexible regulation, financial liquidity and lack of support from the government which are not conducive to the growth of the teak industry in the country. In 2021, the government improved the law related to teak industry and announced the promotion of planting 26 million rai (1 rai = 0.4 acre) of valuable trees and teak (*Tectona grandis* L.f.) is one of the major trees that was promoted. However, with some limitations of teak, such as having a long cutting cycle, makes commercial teak plantations lack of incentives. Therefore, creating channels to

increase income that creates continuous cash flow from non-timber resources with appropriate technology is probably help farmers become more interested in planting economic trees that have a slow cutting cycle. In commercial teak plantation, there will be many seasonal products that have not been systematically used economically such as teak flowers, teak leaves or even the ecosystem of teak forests, etc.

The remaining part of the teak leaves is normally discarded. There are some researches studied the properties of teak leaves and found that teak leaves contain phytochemicals and pharmacological properties that create value of teak leaves greatly. There are a number of studies that have studied extracts from teak leaves (*Tectona grandis* L. f.). It was found that teak contains a variety of bioactive compounds such as flavonoids, alkaloids, tannins, anthraquinones and naphthoquinones and has therapeutic properties including antibacterial, anti-inflammatory, anticancer and antioxidants, etc. and has been popularly used as traditional medicine for a long time. From these properties, it can be seen that teak leaves are ideal for developing health products such as herbal tea. Herbal tea has considered to possess an effect on promoting health and reducing the risk of many types of diseases such as slowing down cell degeneration, reduce blood sugar level, reduce cholesterol levels and relieve diarrhea, etc. Greeshma and Murugan (2017) tested the antioxidant properties by comparing young teak leaves and old teak leaves. The results of the study indicate that both young teak leaves and old teak leaves are composed of various types of phenolic compounds such as gallic, vanillic, p-hydroxybenzoic, ferulic, chlorogenic, sinapic, p-coumarate and cinnamic acids. It was also determined that young teak leaves contain more amounts and types of phenolic compounds than extracts from old teak leaves. Siskawardani and colleagues (2017) reported that teak leaves contain flavonoids, including anthocyanin and its antioxidant properties were increased when 4% citric acid was added to the anthocyanin extraction process. There are only a few studies that mention herbal tea made from teak leaves. They report that the active ingredients from teak leaf herbal tea obtained from baking at 60 °C and then soaking in hot water at 80 °C for 20 minutes were found. That teak leaf tea extract has a high ability to bind DPPH free radicals with an EC<sub>50</sub> value of 28.93 µg/ml (green tea has an EC<sub>50</sub> value of 11.903 µg/ml) and had a Total phenolic amount equal to 295.94 milligrams of catechin per gram of extract. (Total phenolic content of green tea is 421.56 milligrams of catechin per gram of extract). In addition to antioxidants, crude teak leaf extract was also found to have an effect on inhibiting 2 types of enzymes, alpha-amylase and amyloglucosidase, related to diabetes. Therefore, the researchers were convinced that teak leaves have high potential to produce healthy tea that has antioxidants but there are a lot of gaps for developing them into "Herbal tea". By improving the production technology that is appropriate to make herbal tea from teak leaves and create the value of teak leaf, the researchers aimed to study the production process of herbal tea from teak leaves and analyze the antioxidant activity. This would result in the increasing value of non-timber resources (non-timber resources) and increase the attractiveness of farmers to plant economic trees such as teak.

## MATERIAL AND METHODOLOGY

### 1. Preparation of raw materials

Wash and clean the teak leaves, reduced in size and then baked at a temperature of 50 °C for 7 hours according to Bocco et al. (1998). After that, it was finely ground into a powder and packed into a plastic bag then kept at a temperature of 5±2 °C for further analysis.

### 2. Produce antioxidant herbal tea from teak leaves using 2 types of tea processing:

**2.1 Green tea processing:** Leaf stems were cut off from the newly picked teak leaves and reduced to a smaller size then processed to green tea using 2 treatments as below:

Treatment 1: Soak reduced teak leaf in hot water at temperature of 80 °C for 30 seconds to 1 minute, then immerse them in cold water immediately and air dry until damp. Roasted in a roaster at

150 °C until the teak leaves are dry. After that, store in a sealed plastic bag at room temperature for further analysis.

Treatment 2: Roast reduced teak leaves in a roaster at a temperature of 150 °C until the teak leaves are completely dry. After that, store in a sealed plastic bag at room temperature for further analysis.

**2.2 Black tea/red tea processing:** leaf stems was cut off from the newly picked teak leaves and reduce the to a smaller size. Leave them in the well air circulation room for overnight then process to black/red tea using 2 treatments as below:

Treatment 1: Roasted in a roaster at 150 °C until the teak leaves are dry to a certain extent and was roll with a rolling machine until the teak leaves curl into the shape of tea. Leave them for fermentation for 15-30 minutes. Then roasted in a roaster at a temperature of 150 °C to allow the teak leaves to dry completely. After that, they were stored in a sealed plastic bag at room temperature for further analysis.

Treatment 2: Roll them with a rolling machine until the leaves curl up into a tea leaf shape. Leave them for fermentation for 15-30 minutes. Then roasted in a roaster at a temperature of 150 °C until the teak leaves are completely dry and stored them in a sealed plastic bag at room temperature for further analysis.

### 3. Extraction the active substances from teak leaves and teak herbal tea by boiling method

Weigh 30 grams of sample and boil it in hot distilled water (temperature 80-90 °C) in a volume of 300 ml on a hot plate for 30 minutes. Separate the liquid of extracted tea leaf with whatman filter paper no. 1 and concentrate it with a rotary evaporator. Store the extract in refrigerator at 4 °C.

### 4. Antioxidant activity was analyzed using the 2,2 diphenyl-picrylhydrazyl (DPPH) method

Antioxidant activity was tested using the DPPH free radical scavenging activity method adapted from Musa et al. (2013). Weigh 0.00394 g of DPPH solution and dissolve it in 50 mL of EtOH (0.1 mmol). Then dilute the desired sample substance to the following ratios: 1:2, 1:4, 1:8, 1:16, 1:32, 1:64 (equivalent to 50, 25, 12.5, 6.25, 3.125, 1.56 µl/100

µL) in Ethanol, pipette solution into a 100 µl microtiter plate and add 100 µl DPPH solution. Mix the solution well and leave in the dark place for 30 minutes. Pipette into a 100 µl microtiter plate and measure the absorbance at wavelength of 517 nm. Calculate the inhibition (%) using the following formula:

$$\% \text{ inhibition} = (A_0 - A_s) / A_0 \times 100$$

When  $A_0$  = absorbance value of Control

$A_s$  = absorbance value of sample

Calibration curve was performed using the vitamin c equivalent antioxidant capacity (VCEAC) method by preparing L-ascobic acid solutions in Ethanol with concentrations of 1.56, 3.125, 6.25, 12.5, 25, 50, 100 and 1,000 µg/ml.

### 5. Sensory evaluation

Take 3 grams of sample then 150 milliliters of hot water at 100 °C was added. Leave for 3 minutes and separate the tea powder from the liquid. Sensory evaluation was tested using the Hedonic scale scoring test. The panelists gave their satisfaction in the form of a level score. Product likes and dislikes from a given scale a total of 40 untrained panelists. The results of consumer acceptance were then analyzed with the statistical program.

### 6. Statistical data analysis

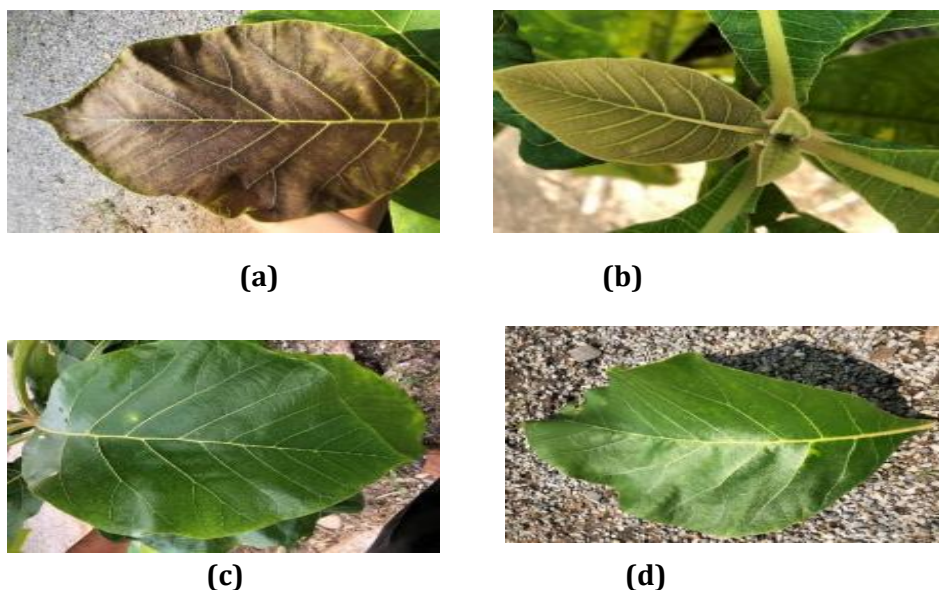
The experiment was proceed using Completely Randomized Design (CRD) and analyze the variance of the data using Analysis of Variance (ANOVA) with the Statistical Analysis System (SAS) program

version 6.12 (SAS Institute Inc, Carry, NC., USA) and compare the means using Duncan's New Multiple Range Test (DMRT) at 95% confidence level ( $p < 0.05$ )

## RESULT AND DISCUSSION

### 1. Sampling and selecting raw materials for tea production

Collected samples of teak leaves were from teak plantations in Nakhon Nayok Province, Thailand. Teak leaf samples was divided into 4 stages: 1. Young leaves (1<sup>st</sup>-2<sup>nd</sup> leaves) 2. Developing leaves (3<sup>th</sup>-4<sup>th</sup> leaves) 3. Fully developed (mature) leaves and 4. Old leaves (as showed in figure 1).



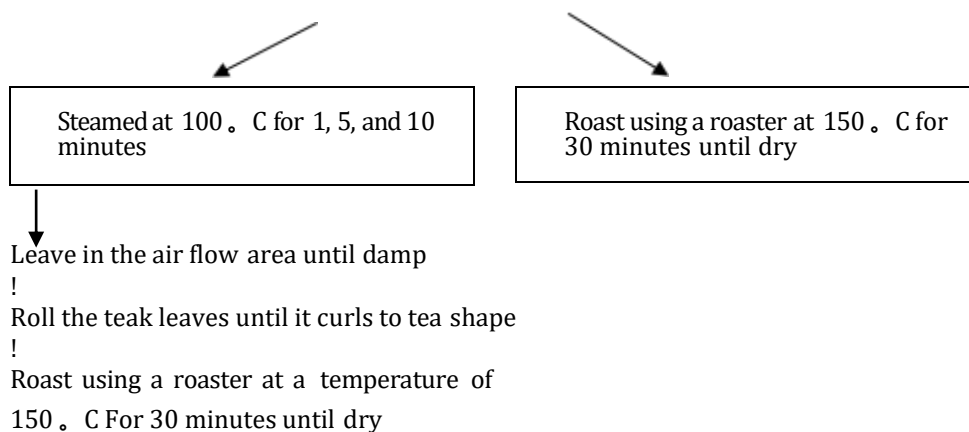
**Figure 1: Characteristics of teak leaves at various growth stages (a) young leaves (stage 1), (b) developing leaves (stage 2), (c) fully developed leaves (stage 3), and (d) Old leaves (stage 4)**

### 2. Herbal tea from teak leaves processing

Teak leaves from stages 1-4 were used for green tea and oolong tea production. The results showed that teak leaves in stages 3-4 could not be used to produce good quality tea (picture not shown) since the leaves are too old and hard resulting uncured and powder-like tea. Therefore, stage 1-2 leaves were chosen to produce tea for the next step.

#### 2.1 Green tea processing from teak leaves

Take the first or second leaf and cut off the stem then cut it into smaller sizes.



**Figure 2: Diagram showing the experiment process of green tea from teak leaves**

From the experiment, it was found that the process of producing green tea through steaming is not suitable because the color of fresh teak leaves (green or purple) dramatically changes to brown by heat (Figure 3).



Steam for 10 minutes



Steam for 5 minutes    Steam for 1 minute    Non-steam

**Figure 3: The color of teak leaves obtained from steaming at 100 . C**

From the results of the product, it was found that steamed teak leaves at 5 and 10 minutes were not suitable for kneading process with the machine (or by hand) because steamed teak leaves are deformed easily, unable to curl and break into powder which are undesired characteristics of tea. Therefore, to process steamed teak leaf required some modification by skipping the rolling process and was immediately roasted in a roaster for 17 minutes. The latter process allowed steamed teak leaf still in shape and slightly curled as shown in figure 4.



**Figure 4: Roast steamed teak leaves for for 17 minutes**

For the second treatment that using non-steamed teak leaf and directly put them in a roaster at a temperature of 150 . C for 20-30 minutes depending on the moisture content until it dry. The resulting showed that the leaf was slightly curl and still in shape similar to regular commercial tea (figure 5).

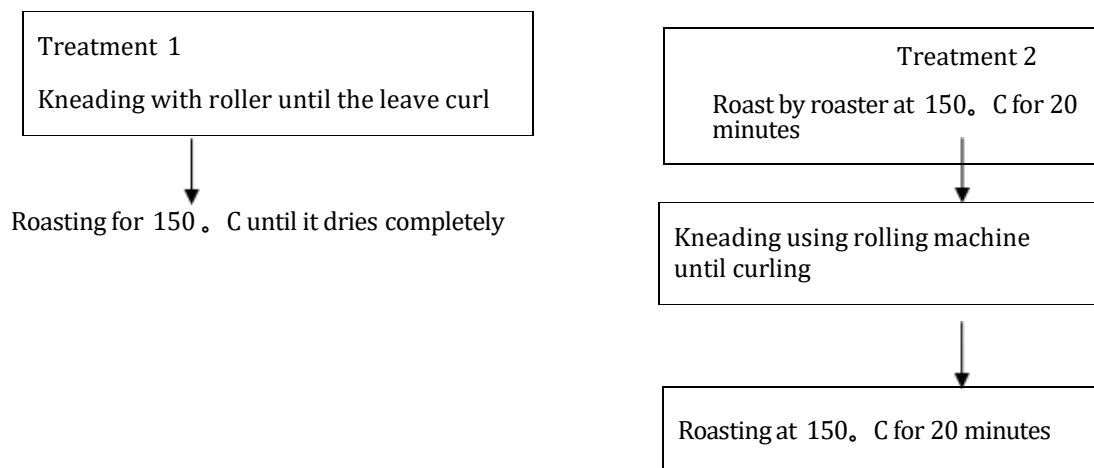




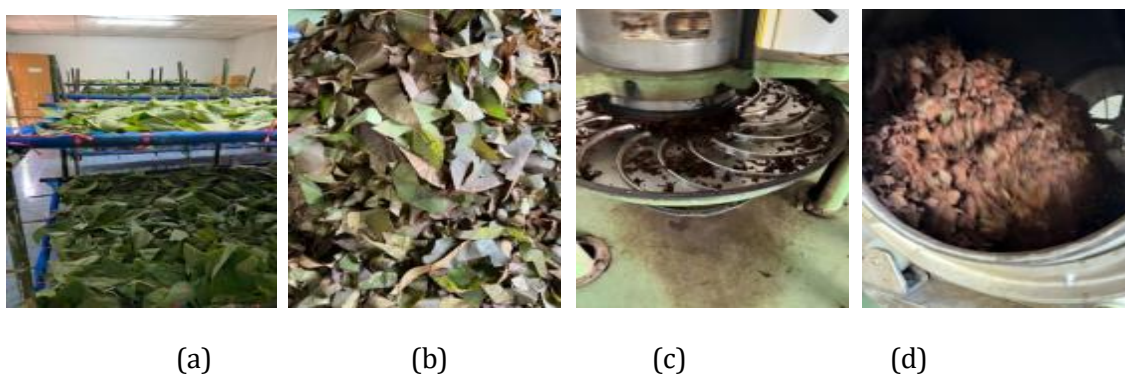
**Figure 5: Green tea produced from non-steamed teak leaves using roasting process**

### 2.2 Oolong tea processing

Take the first or second stage of leaves and cutoff the stem then reduced them into smaller sizes. Drying in the shade for 12 - 16 hours and processed to black tea using 2 treatments as followed:



**Figure 6: Oolong tea processing from teak leaves**



**Figure 7: Oolong tea processing: (a) Dry teak leaf in the shade, (b) Roast until the it withers, (c) Rolling and (d) Drying in the roaster**

From the result, oolong tea from teak leaf is successfully obtained from the traditional protocol with specific condition of drying temperature and period. The characteristic of tea received from the process was comparable to the commercial one with slightly different shape (less curl). Therefore, the existing technology is suitable for making black/red tea from teal leaf with some condition adjustments.

Yield of teak leaf tea is based on the production process of green tea and black/red tea. The results of

each production process are shown in Table 1.

**Table 1: Yield of teak leaf tea obtained from different production processes**

Processing	Treatment	Steps			Initial weight (Kg)	Product weight (Kg)	Yield (%)
		Roasting	Rolling	Drying			
Green tea							
	Air dry						
	Non steam	×	√	√	7.85	3.45	43.95
	Steam 1 minute	×	√	√	6.00	1.80	30.00
	Steam 5 minutes	×	×	√	0.80	0.20	25.00
	Steam 10 minutes	×	×	√	0.40	0.35	87.50
	No air dry	×	×	√	2.35	0.55	23.40
Oolong tea							
	No air dry	√	√	√	9.98	2.25	22.55

From table 1, yield of teak leaf tea was varied from 22.55-87.50 percent depending on the initial moisture content (data not shown) and the process of making tea. The least yield is from oolong tea product with 22.55 percent where the highest yield was obtained from green tea with steaming for 10 minutes. The result was aligned with the other report indicated that the yield of tea production effected from the material itself and the production techniques (Chiu, 1990).

### 3. DPPH free radical scavenging activity analysis

The results of the antioxidant investigation using the DPPH technique and calculate the dilution value of extracted sample of 1 : 16 which the original color of the extract was least disrupted. The half maximal inhibitory concentration (IC<sub>50</sub>), inhibition rate (%) and the amount of antioxidant shown in Table 2.

**Table 2: Percentage of IC<sub>50</sub> inhibition and average amount of antioxidants.**

Processing	Treatment	Steps			Inhibition (%)	IC <sub>50</sub> (μl/mL)	DPPH (μg vitamin c equivalents)
		Roast	Roll	Drying			
Green tea							
	Air dry						
	Non steam	×	√	√	70.04±3.36	2.58	6,542.70±341.15
	Steam 1 minute	×	√	√	72.59±1.76	5.59	6,800.94±178.23
	Steam 5 minutes	×	×	√	77.74±0.17	5.22	7,323.03±16.84
	Steam 10 minutes	×	×	√	73.84±1.08	13.01	6,927.25±109.47
	Steam 20 minutes	×	×	√	67.00±16.05	7.92	6,233.94±1626.96
	No air dry	×	×	√	69.32±9.29	5.38	6,469.72±941.37
Oolong tea							
	No air dry	√	√	√	39.04±3.16	5.99	3,398.94±319.98

From table 2, IC<sub>50</sub> (half-maximal inhibitory concentration) of teak tea was varied from 2.58-13.01 μl/mL which the lowest value is green tea product without steaming. The inhibition rate and the

amount of antioxidant were 39.04-77.74% and 3,398.94-7,323.03 µg vitamin c equivalents, respectively. The inhibition rate of some samples (green teak leaf tea obtained from steaming 5- and 10-minutes process) was comparable to green and black tea report from Moraes-de-Souza et al. (2009). The IC50 value of all samples showed their strong antioxidant efficiency with less than 50 ppm and non-steam green tea was the most effective antioxidant effect with high amount of antioxidant substance and high inhibition rate. The result showed no relationship between IC50 value and percent inhibition which normally the high percentage of inhibition rate would possess low IC50. This might cause from the composition of antioxidant substance presented in the extraction. Too many different types of antioxidants may lead to high inhibition rate with low overall efficiency (Kedare and Singh, 2011). Complexation of reaction between different types of antioxidants and DPPH might occur which consequence an error of analysis (Sharma and Bhat, 2009). This assumption supported by the research of Suryantietal. (2020) found that young teak leaf consists of various antioxidant compounds such as anthocyanins which has maximum absorbance at 208, 489 and 492 nm, 4-hydroxy-4-methyl-2-pentanone, glycerin monoacetate, glycerin diacetate and 1-eicosanol.

#### 4. Sensory evaluation test results

Two selected teak tea samples, according to the production process and free radical activity, were 1) green tea obtained from steaming for 5 minutes follow by roasting (designated as L) and 2) green tea produced from roasting process only (designated as sample F) compared with control sample (untreated teak leaves, in the table designated as sample M). Untrained 42 evaluators were classified as 9 males and 33 females with a wide range of ages from under 20 years old (7 persons) to over 55 years old (4 persons) while the majority of them are 20-25 years old (26 persons). The sensory test was done using a 7-point Hedonic scale scoring test that show the differences of characteristics liking and also help determine the level of product acceptance. The panelists gave their satisfaction in the form of a level score as follows: 1 = dislike very much, 2 = dislike moderately, 3 = dislike slightly, 4 = neither like nor dislike, 5 = like slightly, 6 = like moderately and 7 = like very much. The statistic test results were shown in Tcs4- able 33.

**Table 3: Sensory evaluation of green teak leafherbal tea**

Sample	Color*	aroma	bitterness	turbidity*	Overall liking
L	5.45±1.06	4.81±1.23 <sup>a</sup>	4.36±1.50 <sup>a</sup>	5.74±1.01	4.98±1.02 <sup>a</sup>
F	5.10±1.43	4.83±1.32 <sup>a</sup>	4.74±1.47 <sup>a</sup>	5.74±1.08	5.00±1.29 <sup>a</sup>
M (Control)	5.67±1.16	5.67±1.00 <sup>b</sup>	5.45±1.25 <sup>b</sup>	6.07±1.95	5.83±1.20 <sup>b</sup>

Note: \* means there is no significant difference.

From Table 3, the result showed that sample L and F had sensory evaluation results in terms of color and turbidity that were not different from control samples. While the average values of aroma, bitterness and overall liking were significantly lower than the control sample. The product's liking score for bitterness had the lowest mean value of 4.36 and 4.74 for sample L and F, respectively. This maybe caused by the high amount of total phenolic substances (data not shown), while the turbidity-clearness liking score had the highest value of 5.74 (liking slightly to liking moderately). However, the average score of overall liking was 4.98 (liking it slightly) which can be said that teak leaves can be used to produce herbal tea with a certain level of acceptance of the product by consumers.

#### CONCLUSION

Green and Oolong teak leaf tea was producible from existing tea processing technology with some modification. Steaming step is not suitable for making green teak tea since physicochemical properties of teak leaf is difference from tea leaf which resulting brownish color and unacceptable smell. The antioxidant property of final green tea product is rather similar from control where black



teak tea is much lower. The sensory evaluation of green teak tea showed that the overall liking was liking slightly to liking moderately. The research revealed that teak leaf has potential to be raw material for making antioxidant herbal tea and further study for improving of the flavor is needed.

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