Investigation on The Lifespan of Gaharu Tea

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Abstract— Gaharu tea is a tea that made up from leaves of Aquilaria species. This species has a lot of benefits towards human health such as anti-allergic, anti-diabetic, anti-cancer and anti-inflammatory [1]. In this study, Aquilaria malaccensis has been chosen as the sample due to high distribution of this species in Malaysia. Aquilaria malaccensis is one of the natural species that grow in peninsular Malaysia. This study will investigate the effect of different moisture content and the effect of different storage conditions on the lifespan of gaharu tea. The leaves will be processed using green tea processing method. Based on previous research [5], the drying time is fixed while the temperature and pressure are varied. Therefore, in this research it will undergo drying process at fix temperature (50°C) and pressure (0.6 bar) with different residence time which is 30 mins, 60 mins, 90 mins, 120 mins and 150 mins by using Vacuum Far-Infrared Radiation (VFIR) dryer. After obtained the optimum drying time, the next sample will undergo drying process at fix temperature (50°C), pressure (0.6 bar) and residence time (120 mins) but different storage conditions which is in the kitchen and air conditioning space to determine the optimum lifespan. Lifespan sample will be observed for 4 weeks and the sample will be analyzed every week. All of the samples will be going through moisture content analysis, colony count and chemical analysis. The final result concluded that air conditioning space as the suitable storage condition in order to preserve the nutrient contents.

Keywords— Gaharu tea, Lifespan, Aquilaria Malaccensis

I. INTRODUCTION

Tea is originated from china and it is makes from leaves of plant *Camellia Sinensis*. It is the most popular beverage consumed by two-thirds of the world's population. Since ancient times, tea has been considered a health-promoting habit because of the benefits [2]. Currently, there are few types of tea such as black tea (fermented), green tea (non-fermented) and oolong tea (semi-fermented). These types of tea differ in how the tea is produced and processed based on the different process of drying and fermentation. Black tea is processed by let the tea leaves ferment for several hours before e being either smoke fired, flame fired or steamed. Young tea leaves were used to produced green tea without fermentation after withering, steaming or pan firing, drying and grading. Oolong tea is produced by a partial oxidation of the leaf, intermediate between the process for green and black tea.

As the technology of producing tea is continuously develop, there are other tea that have been produced from different plant such as gaharu tea. The step in producing this tea is just the same as the normal tea. Gaharu tea is made from leaves of Aquilaria species (*Aquilaria malaccensis*) tree. *Aquilaria malaccensis* is one of the expensive trees in the world and this tree mostly used in fragrance industry because of the essential oil from its trunk. Malaysia is one the countries that has high population of this *Aquilaria malaccensis* species other than Thailand, Indonesia, Vietnam and Bangladesh.

Other than its valuable essential gaharu oil, the *Aquilaria malaccensis* leaves consist a lot of benefits. The leaves can be used to make many products such as shampoo, soap and also tea. From previous research finding, many pharmacological activities from the various part of the Aquilaria tree such as Anti-diabetic (anti-hyperglycemic). It can be found at the leaves of Aquilaria spp. The extraction from the leaves consist of methanol extract and it can reduce the blood glucose levels by 40.30% [1]. This research will analyze on the gaharu tea itself because it is the product that need to be consume by our body to gain it benefits so it is important to know the details about the lifespan and the effect on nutrients content.

The critical part in the production of gaharu tea is to determine the lifespan of the tea. Lifespan or shelf life of food is a period for the food to retains an acceptable quality in point of safety and organoleptic point of view [3]. There are four factors that effects the lifespan namely formulation, processing, packaging and storage conditions. In this research, focus will be on storage conditions. Experiments will be done on 2 storage conditions which is kitchen and air conditioning space. These storage conditions have been chosen due to the tea usually placed to keep at house (kitchen) and office (air conditioning). Therefore, it is crucial to know the different and effect of these storage conditions towards the gaharu tea.

Further research will be conduct to investigate the effect of moisture content removal, colony count and antioxidant on the lifespan of Gaharu (*Aquilaria malaccensis*) tea.

II. METHODOLOGY

A. Materials

Fresh leaves of *Aquilaria malaccensis* were taken from farm at Jalan Kebun, Shah Alam. Only good leaves were chosen and damage leaves were avoided to sustain the quality of tea. The leaves were rinsed and wiped using tissue to remove the dirt. Then, keep it inside vacuum seal bag and stored inside refrigerator.

B. Experimental Procedure

i. Drying Process

The leaves undergo drying process using VFIR by controlling drying time and fix the temperature and pressure at 50°C and 0.6 bar respectively. There are 16 leaves that has been arranged on the tray before put it in the VFIR for each drying time. The drying time varies at 60 mins, 90 mins, 120 mins, 150 mins, and 180 mins. After the drying process, the leaves were grounded until it become small particle using dry mill. The grounded leaves were put it in the vacuum seal bag and stored inside desiccator containing silica gel before proceed with the analysis.

For lifespan experimental procedure, 120 minutes has been chosen as optimum drying time. 16 clean leaves were placed on the tray for drying using VFIR. After drying, the dried leaves were grounded using dry mill until it become small particle. Then, the grounded leaves were weighed for 2g using weighing balance. the 2g of grounded leaves has been placed inside the tea bag and store it inside a plastic container. The steps were repeated until got 20 samples. The sample were divided into 2 storage condition which is kitchen and air conditioning space. The plastic container containing sample has been placed to this both places.

ii. Moisture Content Removal

The moisture analysis was conducted using Sartorius Infrared Moisture Analyzer MA35. The temperature and time were set up for 100°C and 5 min respectively. Put 2g of sample inside the moisture analyzer and let it run. This analysis is to see the quantity of moisture content removal between different drying time. The result was recorded.

Lifespan analysis used same setting for moisture content removal. 2g of sample inside tea bag from each storage condition was picked and put inside the moisture analyzer one by one. The result was recorded.

iii. Colony Count

Nutrient agar was prepared by mix 20g of nutrient powder with 1000ml of distilled water. Then, put the solution inside the autoclave to sterilized at 120°C for 15 minutes. After that, take out the solution and let it cool down to room temperature. Poured the solution into the petri dish and keep it inside biological safety cabinet until it solidified. Stored it inside 4°C refrigerator.

The grounded leaves inside the tea bag were swabbed with cotton bud and spread on the nutrient agar to check the colony count for each different drying time sample. The result was checked the next 2 day using Stuart Colony Counter. The data was recorded and for the lifespan procedure, the analysis was conducted every week for 4 weeks.

iv. DPPH (2,2-diphenyl-1-picrylhydrazy) radical scavenging activity

DPPH solution was prepared by mix 7.9mg with 100ml of methanol inside the conical flask. Then, wrapped with aluminium foil and stored it inside the refrigerator with temperature 4°C.

Tea extraction was prepared by immersed 2g of grounded leaves into 200ml of boiling water with temperature 100°C and let it cool to room temperature.

Briefly, 3 ml of each sample tea extraction was reacted with 3 ml of 0.2mM DPPH in 95% methanol inside small bottle sample. Then, the solution was incubated in the dark space at room temperature for 30 minutes. After that the absorbance (A) was measured using spectrophotometer at 517 nm single wavelength [4]. The DPPH percentage was calculated using following equation:

Free radical (%) = A (blank) – A (extract) / A (blank) \times 100

III. RESULTS AND DISCUSSION

A. Determination of optimum drying time

The optimum drying time has been chosen by analyze the moisture content removal, colony count and antioxidant by using DPPH assay.

i. Moisture content removal analysis



Figure 1: Sartorius Infrared Moisture Analyzer MA35

Moisture content removal vs drying time 12 9.05 % 10 8.14 7 89 Moisture content removal 8 6 4 2 0 90 mins 120 mins 150 mins 180 mins 60 mins Drying time (minutes)

Figure 2: Moisture content removal base on different drying time.

Figure 2 show moisture content removal from dried leaves after drying process using VFIR. The leaves has lost some weight due to moisture loss from the leaves itself. Drying is a method to preserve the food by evaporate the moisture inside the leaves. Base on previous research [5], the optimum condition obtained is 50°C and 0.6 bar at constant time. In this research the drying time has been varying 60 minutes. 90 minutes, 120 minutes, 150 minutes and 180 minutes to study the optimum drying time. From figure 1 show that at 120 minutes the percentage value is 8.14% and the value is close to the previous research which is 8.55%.

ii. Colony count analysis

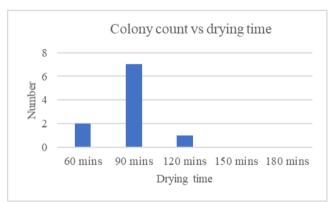


Figure 3: Number of colonies on the agar plate.

Colony count is a method that use nutrient agar to check the present of bacteria on the food. Figure 3 show the number of colony on the agar plate after 2 days. It show the decreasing on colony number when the time increase eventhough on 90 minutes the colony number is higher. Bacteria is highly attracted to moist condition of food, because of that there is high colony count in the beginning cause the shorter drying time.

iii. DPPH Assay

Table 1: Antioxidant activity of gaharu tea extraction.							
Time	60	90	120	150	180		
(minutes)							
DPPH	41.9±0.12	81.1±0.09	86.4±0.19	84.9 ± 0.070	93.4±0.13		

Based on table 1, 180 minutes has the highest antioxidant with 93.4% compared to other drying time. The high the percentage value, the higher the antioxidant activity [6]. Antioxidant play an important role in our body, it helps toprevent the formation of free radicals and delay the development of lipid peroxidation [7]. Free radicals can cause damage to cellular protein and eventually cell death [8].

Optimum drying time have been chosen by analysed all the data obtained. It also continuity base on previous research [5]. The chosen drying time for *Aquilaria malaccensis* leaves is 120 minutes. The value of antioxidant is higher at 180 minutes however

it cannot be choose as optimum drying time. This selection has been made by look at the moisture removal which has the closest value with the previous research [5] and the colony count analysis result showed that at 120 minutes there is only one colony on the agar plate and that is meant that the drying time can disintegrate the bacteria. Eventhough there is zero colony at 150 minutes and 180 minutes but the selection is base on overall analysis, so drying time 120 minutes have been chosesn due to most good compatibility.

B. Gaharu tea lifespan analysis

i. Moisture content removal analysis

For lifespan analysis, gaharu tea sample has been placed at two different storage condition. One sample is place in the kitchen while the other one in air conditioning space. This storage condition has been chosen because it is the usual place to kept and store the tea at home or at the office.

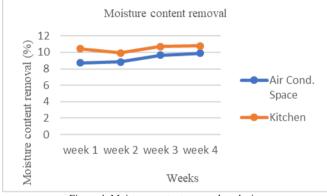


Figure 4: Moisture content removal analysis.

Figure 4 show the moisture content removal for kitchen and air conditioning storage condition. Based on observation from the graph, it showed the increasing of trendline along with the increasing of storage time. Furthermore, the value of moisture removal from kitchen is higher than air conditioning space.

As stated in figure 4, the value of kitchen moisture removal is more than 10% and this meant there is more moisture loss from the leaves. While for air conditioning space, the value is below 10% event though the trendline is increasing. In research by K. Brown [9], it stated that low humidity level can increase product water loss and this explain why moisture removal value in the kitchen is higher since it is low humidity level compared to the air conditioning space.

The humidity in the air conditioning space is high due to the cool air release from the air conditioner. Generally, water osmosis will move from high to low and in this situation high humidity in the air conditioning space will prevent the moisture from the leaves to move out because the humidity is already high.

ii. Colony count analysis

This method is a simple method and it is faster and cheaper than conventional plating method. As mention earlier this analysis is to determine the present of bacteria on the sample throughout the 4 weeks at 2 different storage condition.

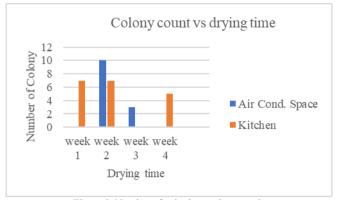


Figure 5: Number of colonies on the agar plate.

Figure 5 show the result of colony count after 2 days inside the incubator at temperature 37°C. As stated on the figure, the trend is not consistent and this may be due to a few factors. Theoretically, tea inside a tea bag have high probability to be contaminated. In addition, it was found that pathogen on contaminated tea increase significantly over time [10].

Therefore, it is probably that this sample have been contaminated along the preparation which is one of the factors that cause the inconsistent results. Besides, it may be due to poor handling during the experiment. However, it still showed that air conditioning space is much better because there is no colony count on week 1 and 4 while for the kitchen sample only week 3 that have zero colony count.

iii. DPPH assay

Table 2: Antioxidant activity of gaharu tea extraction for 4 weeks							
Time	1	2	3	4			
(weeks)							
DPPH							
(1)	88.5 ± 0	87.3±0.03	92.4±0.09	87.1±0.12			
DPPH							
(2)	88.3±0.09	90.0±0.03	90±0.03	87.9±1E-14			
$(1) K' \in I (2) A' = I' : : :$							

(1): Kitchen, (2): Air conditioning space

Based on table 2, percentage of antioxidant activity lowest on week 4 for kitchen condition with only 87.1% while for air conditioning space with 87.9%. The value likely constant on week 2 and 4 but high on week 3 for kitchen condition. However, overall air conditioning space has slightly higher and stable antioxidant activity compared to kitchen with highest value 90% on week 2 and 3 while the highest value for kitchen only on week 3 which is 92.4%. Therefore, air conditioning space is much more suitable storage condition to keep the gaharu tea in terms of antioxidant.



Figure 6: tea extract using water extraction (air conditioning space)



Figure 7: tea extract using water extraction (kitchen)

IV. CONCLUSION

Gaharu tea has many benefits for the consumer and it is also knowing as one of herbal tea. In order to keep and store the gaharu tea leaves, it must in suitable area to avoid from nutrient lose. As conclusion, air conditioning space is a suitable place to store the tea base on the discussion. This is supported by Selena Ahmed in her article that said it is preferable to keep and store the tea in seal packaging, cool and dark conditions in order to extend the shelf life [11].

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