The utilization of Sambung Nyawa (*Gynura procumbent* [Lour. Merr]) leaves extracts as an antioxidant for coconut oil by using ethanol solvent

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The utilization of Sambung Nyawa (*Gynura procumbent* [Lour]. Merr) leaves extracts as an antioxidant for coconut oil by using ethanol solvent

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**Abstract.** Sambung Nyawa (*Gynura procumbent* [Lour]. Merr) is commonly well-known as traditional medicine. It contains several chemical constituents such as flavonoids, saponins, tannins and steroids or triterpenoids, which are potential as an antioxidant. The aim of this study was to analyze the effect of Sambung Nyawa leaves to extract as an antioxidant and its ability to preserve the quality of coconut oil. First, flavonoids were extracted from Sambung Nyawa leaves at various leaf to the solvent ratio (w/v) and extraction temperature. The extracts which gave the highest total flavonoids content was added into the coconut oil and stored for 3 days, 6 days, 9 days, 12 days, and 15 days. Total flavonoids content of Sambung Nyawa leaves extracts were evaluated by UV-Vis spectrophotometry. Coconut oil was analyzed for its value of acid, iodine, and peroxide number. The results showed that the highest total flavonoids content of 5.18% was obtained for the leaf to solvent ratio of 1:15 (w/v) and an extraction temperature of 65 °C. The lowest acid number of 0.35%, the highest iodine number of 8.09 g I$_2$/100 g, and the lowest peroxide number of 5.20 mg O$_2$/100 g was obtained for the storage time of 3 days for coconut oil mixed with the Sambung Nyawa leaves extracts.

1. Introduction

The plant of Sambung nyawa is a traditional herb comes from South East Asia that contains several active chemical constituents such as flavonoids, saponins, tannins, and steroids or triterpenoids [1]. The plant has anti-Hyperglycemic, anti-Hyperlipidemic, analgesic [2]. The classification of Sambung Nyawa is shown in Table 1.

| Table 1. The Classification of Sambung Nyawa | Spermatophyta |
|---------------------------------------------|---------------|
| Division                                     | Spermatophyta |
| Subdivision                                  | Angiospermae  |
| Class                                        | Dicotyledoneae|
| Order                                       | Asterales     |
| Family                                      | Asteraceae    |
| Genus                                       | Gynura        |

Coconut oil is a vegetable oil that produced from coconut meat. The fatty acids found in coconut oil consist of 90% saturated fatty acids and 10% unsaturated fatty acids [3]. The quality standards of coconut oil are 5% of maximum acid number, 8-10 g I$_2$/100g of iodine number and 5 mg O$_2$/100 g of maximum peroxide number [4]. Lots of treatments have been done to improve the quality of coconut
oil as the development of technology, such as an addition of antioxidants into coconut oil to prevent oxidation [5]. Coconut oil oxidation may occur due to contact between the amount of oxygen and the coconut oil and the heating process [6].

Antioxidants are synthetic or natural. The function of the substances that may delay, retard or prevent the process of oxidation. Based on the research, synthetic antioxidants turned out to be carcinogenic, so that the usage of synthetic antioxidant began to be restricted [7, 8]. Natural antioxidants are commonly known as phenolic or polyphenolic compounds, such as flavonoids, cinnamic acid derivatives, coumarin, tocopherol, and polyfunctional acids [9]. Flavonoids are the largest phenolic group found in nature [10]. Flavonoids are widely distributed in all parts of the plant including fruit, root, leaf and stem epidermis. Flavonoids have been reported to have antioxidant activity which can inhibit the cell damage caused by oxidation [11, 12]. Flavonoids have the ability to change or reduce free radicals and also as anti-free radicals [8]. This extraction method is the most common method and easy to use to obtain the active substances [13].

In this study, flavonoids were extracted from Sambung Nyawa leaves by using ethanol solvent and reflux extraction method by varying temperature time and the ratio of raw material and solvent. The effectiveness of flavonoids contained in Sambung Nyawa leaves extracts as an antioxidant for coconut oil will be evaluated.

2. Methods

2.1. Materials
Sambung Nyawa leaves were collected from Medan, Indonesia. Ethanol 96% was produced by Rovin Chemical Indonesia.

2.2. Preparation of Sambung Nyawa Leaves Powder
Sambung Nyawa leaves were washed using clean water and left for 3-4 days (no solar heat) until completely dry. The dried leaves were blended into powder using a blender and then they were sieved using a strainer of 40 mesh/inch to equalize the size.

2.3. Flavonoids Extraction from Sambung Nyawa Leaves Procedure of Powder
Extraction of flavonoids was done by using reflux extraction method and ethanol as solvent. Sambung Nyawa leaves powder (25 grams) was mixed with (1:5; 1:10; 1:15) (w/v) of ethanol 96% and heated using a hot plate (35 °C, 45 °C, 55 °C, 65 °C) for 3 hours with 300 rpm stirring. The extracts were filtered using Whatman no.1 filter paper and then heated in the water bath (50 °C) for the viscous extracts. Total flavonoids content was analyzed by using UV-Vis Spectrophotometry in BTKLPP Medan.

2.4. Procedure of The Utilization of Sambung Nyawa Leaves Extract as an Antioxidant
Coconut oil (15 grams) was mixed with Sambung Nyawa leaves extract (5% of coconut oil mass) and put into the black bottle and stored for (3 days, 6 days, 9 days, 12 days, 15 days). The effectiveness of flavonoids contain in Sambung Nyawa leaves extracts was evaluated based on acid, iodine and peroxide number of coconut oil. The analysis were done for the coconut oil mixed with Sambung Nyawa leaves extracts, and the coconut oil with no addition of Sambung Nyawa leaves extract. The standard that used for the coconut oil characteristics based on SNI 01-2902-1992.

3. Results and Discussion

3.1 Total Flavonoids Content in Sambung Nyawa Leaves Extracts
Figure 1. The effect of material to solvent ratio (w/v) and Temperature extraction to total flavonoids content (%)

Figure 1 showed the effect of material to the solvent ratio (w/v) and extraction temperature to total flavonoids content (%). For the ratio of 1:5 (w/v), total flavonoids content increased with increasing the extraction temperature. Total flavonoids content of 1.50%; 1.53%; 1.59%; 2.07% were obtained at 35 °C, 45 °C, 55 °C, 65 °C, respectively. The ratio of 1:10 (w/v) gave the fluctuation total flavonoids content. Total flavonoids content of 1.79%; 2.76%; 2.09%; 3.30% were obtained at 35 °C, 45 °C, 55 °C, 65 °C, respectively. For the ratio of 1:15 (w/v), total flavonoids content increased with increasing the extraction temperature. Total flavonoids content of 1.90%; 2.90%; 3.33%; and 5.18% were obtained at 35 °C, 45 °C, 55 °C, 65 °C, respectively. The extraction temperature and ratio of material to solvent (w/v) are factors that can influence the extraction process. The increasing of extraction temperature caused the movement of molecules of ethanol as solvent random and faster. The pores of the solid of Sambung Nyawa leaves powder swelled. Hence, solvent easier to diffuse into the pores of the solid of Sambung Nyawa leaves powder and dissolved flavonoids compound. So, the rate of extraction was higher and gave higher results. Similarly to the ratio of material and solvent (w/v) that the increase of material to the solvent ratio (w/v) will give the higher results, because the extracted material will contact more often with the solvent which more numerous than the solvent of small amounts [14, 15].

Overall, the results of this study showed that the increase of material to the solvent ratio (w/v) and extraction temperature gave the increase of total flavonoids content (%). But, in this study, there is the deviation for the ratio of 1:10 (w/v) at an extraction temperature of 55 °C where total flavonoids content decreased. It was caused by the presence of impurities in Sambung Nyawa leaves extracts which caused the change of flavonoids characteristic. Hence, the number of total flavonoids content became smaller. The highest total flavonoids content of 5.18% was obtained at temperature extraction of 65 °C and material to solvent ratio of 1:15 (w/v).

3.2 Acid Number of Coconut Oil Analysis

Figure 2 showed the analysis results of an acid number of coconut oil with and without extracts addition. From figure 2, we can see that an acid number of coconut oil increased with the duration of storage time. But, an acid number for coconut oil with extracts addition was lower than coconut oil without extracts addition. The acid number of 0.50%; 0.70%; 1.45%; 1.50% and 1.70% were obtained for coconut oil without extracts addition with storage time of 3 days, 6 days, 9 days, 12 days, and 15 days, respectively. The acid number of 0.35%; 0.55%; 0.90%; 1.25% and 1.45% were obtained for coconut oil with extracts addition with storage time of 3 days, 6 days, 9 days, 12 days, and 15 days, respectively. The lower value of the acid number for coconut oil with extracts addition showed the presence of flavonoids compound in Sambung Nyawa leaves extracts that acted as an antioxidant for coconut oil. Free fatty acid was found in oil or fat since the material was harvested and the amount of free fatty acid will increase during the processing due to the oxidation process that caused the amount
of free fatty acid increased and the value of acid number became higher [3]. According to SNI 01-2902-1992, the maximum value of acid number for coconut oil is 5%. The results of this study gave the value of acid number that fulfill the standard of coconut oil for storage time of 3 days, 6 days, 9 days, 12 days, and 15 days. The lowest acid number of 0.35% was obtained for coconut oil with extracts addition and storage time of 3 days.

**Figure 2.** The effect of extract addition and storage time to acid number of coconut oil

3.3 Iodine Number of Coconut Oil Analysis

Figure 3 showed the analysis results of iodine number of coconut oil with and without extracts addition. From figure 3 we can see that the iodine number decreased with the longer of storage time. But, the iodine number for coconut oil with extracts addition was higher than coconut oil without extracts addition. The iodine number of 7.61 g I₂/100 g; 7.30 g I₂/100 g; 6.73 g I₂/100 g; 6.25 g I₂/100 g and 5.96 g I₂/100 g were obtained for coconut oil without extracts addition with storage time of 3 days, 6 days, 9 days, 12 days, and 15 days, respectively. The iodine number were obtained for coconut oil with extracts addition with storage time of 3 days, 6 days, 9 days, 12 days, and 15 days, respectively. Iodine number indicates the amount of unsaturation fatty acid in the coconut oil. The higher value of the iodine number indicates the better quality of coconut oil. The iodine number can decrease due to rupture of unsaturated bond become saturated caused by the upsurge of oxidation period hence the degree of unsaturation fatty acid in coconut oil becomes lower [16, 17]. Addition of antioxidant into coconut oil inhibits the oxygen bind to the double bonds of fatty acid and delay the oxidation process due to the lower of the amount of oxygen that bind to the double bonds, hence the
iodine number is increasing [18]. Addition of Sambung Nyawa leaves extracts into coconut oil gave the lower iodine number decreasing than coconut oil without Sambung Nyawa leaves extracts addition. According to SNI 01-2902-1992, the value of iodine number was 8-10 g I$_2$/100 g. Coconut oil with extracts addition with storage time of 3 days fulfill the standards of coconut oil, but for the storage time of 6 days, 9 days, 12 days, and 15 days, the iodine number decreased so that the value of iodine number of coconut oil lower than the minimum limit of the standard and for the coconut oil without extracts addition, the value of iodine number didn’t fulfill the range of the standard with storage time of 3 days, 6 days, 9 days, 12 days and 15 days. The highest iodine number of 8.09 g I$_2$/100 g was obtained for coconut oil with extracts addition.

3.4 Peroxide Number of Coconut Oil Analysis

![Figure 4](image_url)

**Figure 4.** The effect of extract addition and storage time to peroxide number of coconut oil.

Figure 4 showed the analysis results of peroxide number of coconut oil with and without extracts addition. From figure 4 we can see that the peroxide number will increase days by days. But, the peroxide number of coconut oil with extracts addition was lower than coconut oil without extracts addition. The peroxide number of 5.60 mg O$_2$/100g; 7.20 mg O$_2$/100g; 8.00 mg O$_2$/100g; 8.40 mg O$_2$/100g; and 10.00 mg O$_2$/100g were obtained for coconut oil without extracts addition. The peroxide number of 5.20 mg O$_2$/100g; 6.00 mg O$_2$/100g; 6.40 mg O$_2$/100g; 6.80 mg O$_2$/100g; and 8.40 mg O$_2$/100g were obtained for coconut oil with extracts addition. The peroxide number showed the rancidity of coconut oil due to oxidation and hydrolysis. The high peroxide number indicates the fat or oil has oxidized. The peroxide number of coconut oil will increase along with the duration of the storage time [6, 18]. Coconut oil with extracts addition gave the lower peroxide number than coconut oil without extract addition. According to SNI 01-2902-1992, the maximum value of peroxide number was 5.0 mg O$_2$/100 g. The results of peroxide number analysis showed the value that higher than the maximum limit of standard with storage time of 3 days, 6 days, 9 days, 12 days, and 15 days. The lowest peroxide number of 5.20 mg O$_2$/100g was obtained for the coconut oil with extracts addition and storage time of 3 days.

4. Conclusion

In this study, the highest total flavonoids content of 5.18% that was obtained at the extraction temperature of 65 °C and material to solvent ratio of 1:15 (w/v) was added into coconut oil. To evaluate the flavonoid activity in Sambung Nyawa leaves extracts as an antioxidant, the coconut oil with extracts addition was compared to coconut oil without extract addition based on some characteristics of coconut oil which included the acid number, iodine number, and peroxide number. The results showed that the lowest acid, the highest iodine number, and the lowest peroxide number of 0.35%; 8.09 g I$_2$/100 g and 5.20 mg O$_2$/100, respectively were obtained for coconut oil with extracts addition and storage time of 3 days. Sambung Nyawa leaves extracts that was added into coconut oil
can retard the oxidation process, hence gave the better characteristics than coconut oil without extracts addition.

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