Original Article
Food Science and Technology

ISSN 0101-2061 (Print)
ISSN 1678-457X (Online)

DOI: https://doi.org/10.5327/fst.00421

Quality of sengkubak leaf simplicia from various stages of maturity and drying methods

Bagem Br SEMBIRING[1,2], Muhammad Ali FULAZZAKY[2,3], Helmi HARIS[2], Ridwan RACHMAT[4*], Dian HISTIFARINA[4], Tika Hafzara SIREGAR[1], Agus TRIYONO[4]

Abstract

The sengkubak plant, especially its young leaves, has been used by the Dayak tribe as a food flavoring for generations. Apart from being a flavoring, it can also be a medicinal ingredient. The sengkubak plant has not been cultivated; it grows wild in the forest. The research aims to determine the effect of the drying method and the level of leaf maturity in producing quality simplicia with broad benefits. Parameters consisted of dry weight yield, moisture content, color, protein content, antioxidant activity, and leaf chemical compounds. The highest dry weight yield of sengkubak leaves ranged from 25.94 to 27.10%, the lowest moisture content was 6.79–7.05%, and the smallest IC_{50} (inhibition concentration 50%) value, namely, 249.19–250.50 ppm, was found in the oven drying method. The highest protein content ranged from 20.94 to 22.45% in young and medium leaves. The results of chemical components in sengkubak leaves: the wind-drying method detected 21 chemical compounds in young leaves, 21 in mature, and 17 in old leaves. The method para-para identified 11 components in young leaves, 15 in mature, and 24 in old leaves. Meanwhile, with the oven, young leaves had 15 chemical compounds, medium had 17, and old leaves had 12 chemical compounds. The level of leaf maturity and drying techniques influence the quality of sengkubak leaf simplicia. Quality sengkubak leaves are young and mature that are air-dried and placed on a rack covered with a black cloth under the sunlight. The scanning electron microscopy results show that the powder morphology of young and old leaves is round and hollow, while mature leaves are rod-shaped. Meanwhile, the thermogravimetric analysis results show that the heating temperature that can leave around 50% of the sample is around 364.04–370.29°C with a heating time of around 33.90–34.52 min; there is no difference between young, medium, and old leaves.

Keywords: drying; leaf maturity level; *Pycnarrhena cauliflora*; quality; simplicial.

Practical Application: Practical application of Sengkubak (Pycnarrhena cauliflora) are known to have various benefits, especially as traditional medicine and cooking spices. This research can be used in drying herbal tea products, cooking spices, and extracts of Sengkubak.

1 INTRODUCTION

The sengkubak plant (*Pycnarrhena cauliflora*) empirically and according to the experience of the Dayak tribe can be used as a food flavoring ingredient and also as a medicine for headaches. The young leaves are pounded or chopped and then added to dishes to add a savory and slightly sweet taste to the dish (Juita et al., 2015). According to Tritisari et al. (2023), using powder from sengkubak leaf juice gives a more umami taste and aroma to dishes compared to powder from simplicia. Apart from being a food flavoring, sengkubak leaves are useful as an antioxidant, anti-cancer, anti-acne, anti-bacterial, anti-dental decay, and as a functional food because they contain amino acids and minerals (Masriani & Sunarti, 2019; Purba et al., 2014; Puspita & Wulandari, 2020). The secondary metabolites of sengkubak leaves include alkaloids, flavonoids, triterpenoids/steroids, phenols, and tannins. Sengkubak leaf extract is relatively non-toxic at the test limit of 2,000–5,000 mg/kg body weight (Pamuji, 2015).

Sengkubak plants are not widely cultivated, as much as 71.42% of their growing habitat is still in the forests of the Kalimantan region (Juita et al., 2015). Sengkubak plants grow vines around tall trees. To get fresh leaves, Dayak people go to the forest to pick young leaves. Using fresh leaves takes time because they have to be picked first. Fresh leaves cannot be stored, so they are picked continuously. To meet needs and reduce forest damage, leaves are preserved through a drying process. According to Fahmi et al. (2019), drying is one way to maintain the quality of the material. Sengkubak leaves with various levels of maturity are dried using several drying methods to obtain a drying method and level of leaf maturity that can produce quality simplicia with an umami taste that is no different from fresh leaves and has wider benefits. The drying method has an important role in the effectiveness of drying performance and better quality of the resulting dry product.

Received: Oct. 30, 2024.
Accepted: Dec. 11, 2024.
[1]*National Research and Innovation Agency, Research Center for Agroindustry, Cibinong, Bogor, Indonesia.*
[2]*University of Djuanda, School of Postgraduate Studies, Jalan Tol Ciawi No. 1, Ciawi, Bogor, Indonesia.*
[3]*Universitas Ibn Khaldun, Faculty of Engineering, Department of Civil Engineering, Jalan Sholeh Iskandar, Kedungbadak, Bogor, Indonesia.*
[4]*National Research and Innovation Agency, Research Center for Appropriate Technology, Bogor, Indonesia.*
*Corresponding author: ridw011@brin.go.id
Conflict of interest: nothing to declare.
Funding: none.

2 MATERIALS AND METHODS

2.1 Materials

The research raw material was sengkubak leaves obtained from Upit Village, Belimbingan District, Melawi, West Kalimantan, and the chemical materials consisted of distilled water, 2,2-diphenyl-1picrylhydrazyl (D913-2), methanol pa (EMSURE, ACS, ISO, Reag. Ph Eur), concentrated sulfuric acid, selenium mixture, NaOH 40%, boric acid 1% and 0.05N sulfuric acid, and Whatman filter paper no. 41. Meanwhile, the equipment used consisted of an oven, blender/hammer mill, ultraviolet–visible (UV–Vis) spectrophotometer, titration, and gas chromatography-mass spectrometry (GC-MS). The quality of sengkubak leaf simplicia was analyzed with JEOL JSM-IT 200 scanning electron microscopy (SEM) and a Simultaneous Thermal Analyzer (Model STA 6000).

2.2 Methods

The research was carried out at a Testing Laboratory of the Spice and Medicinal Plant Research Institute, Bogor and Agro-medical Industrial Engineering Development Laboratory, National Research and Innovation Agency, Serpong. The research used a factorial complete randomized design consisting of two factors, namely, the level of leaf maturity (A), young leaves (a1), mature leaves (a2), old leaves (a3), and drying method (B), air drying (b1), racks covered with a black cloth under sunlight (b2), and open oven (natural gas) (b3). Each treatment was repeated three times. The research observation parameters consist of dry weight yield, water content (distillation/toluent), color, protein content (digestion), antioxidant activity with 2,2-diphenyl-1-picrylhydrazyl (DPPH) method, chemical components, SEM analysis, and thermogravimetric analysis (TGA).

2.3 Research procedure

2.3.1 Handling sengkubak leaves

The harvested sengkubak leaves were sorted to separate the leaves according to the level of maturity (young, medium, and old), rotten leaves or those affected by disease/pests, and yellow leaves that were present at the time of harvest. The leaves sorted based on maturity were washed using clean water in a running water area or a washing tub and carried out until they were clean. After washing, the leaves were drained on a draining rack made of wire or a porous plastic basket in a shady place until the water no longer drains.

2.3.2 Drying

Sengkubak leaves were selected according to their maturity level, weighed, and then dried using various dryer techniques according to the treatment. Each drying technique was equipped with a data logger device, which was placed on top of the material to record temperature and humidity during the drying process. After drying, each leaf was weighed for dry weight and then ground separately using a hammer mill to obtain sengkubak leaf powder measuring 50–60 mesh.

2.4 Analysis of the quality of sengkubak leaf simplicial

2.4.1 Protein

Notably, 2 g of sengkubak leaf powder was weighed into a digestion tube, and then 5 ml of concentrated sulfuric acid and 1 g of selenium mixture were added. The mixture was stirred until homogeneous and then left overnight. Next, digestion was carried out at 300°C until the solution became clear. After digestion, dilution was performed by adding distilled water to a 100 ml measuring flask. A volume of 10 ml of the solution was pipetted into a Kjeldahl flask, and 50 ml of distilled water was added. Then, 10 mL of 40% NaOH was added for distillation and collected in a flask containing 10 ml of boric acid. The indicator solution was then added into the flask until the color of the solution changed from pink to green, and then left undisturbed for 10 min. Next, titration was carried out with 0.05 N sulfuric acid until the color of the solution changed to green.

2.4.2 Antioxidant activity

Dried sengkubak leaves were ground into 50 mesh powder. One gram of powder was weighed into an Erlenmeyer flask, and methanol solvent was added, stirred, left to sit, and then filtered. The solution was tested for antioxidant activity using the DPPH (2,2-diphenyl-1 picrylhydrazyl) method. The concentration of the extract solution that had been made was pipetted to 0.5, 1, 1.5, 2, and 2.5 mL. Each was diluted with methanol:water (1:1) in a 10 mL volumetric flask to the limit mark to obtain concentrations of 50, 100, 150, 200, and 250 µg/mL. According to the concentration, 1 mL of the solution was pipetted into a test tube, and 1 mL of 100 µg/mL DPPH solution and 2 mL of methanol were added. The mixture was homogenized and then left for 30 min in a dark place, and the absorbance of the solution was measured using a UV-Vis spectrophotometer at a wavelength of 517 nm. The effectiveness of antioxidants was determined by the IC_{50} value (inhibition concentration 50%). The IC_{50} value indicates the concentration of the sample that can capture 50% of free radicals, as shown by the linear regression line equation, which states the relationship between the concentration of the sample compound (X) and the average antioxidant activity/inhibition (Y) (Equation 1).

$$Inhibition\ concentration\ (IC_{50}) : \frac{A\ Control - A\ Sample}{A\ Sample} \times 100\% \tag{1}$$

2.4.3 Chemical component

Dried sengkubak leaves of various stages of maturity, resulting from drying using some drying methods, were each ground to obtain a 50-mesh powder. The powder was weighed 1 g each into a 50 mL measuring cup, and 96% ethanol solvent was added, stirred, and left for about 1 h. The results of the maceration were filtered using Whatman paper, and the filtrate obtained was ready to be identified using the GC-MS method to determine the chemical components contained in the sample, which was carried out at the LABKESDA of DKI Jakarta, in Rawasari.

2.4.4 The analysis scanning electron microscopy and TGA

The morphological structure of the powder was observed using a JEOL JSM IT 200 SEM. The sample was coated with gold through a coating process with a maximum thickness of 10 µm so that the surface shape of the sample did not change (Boldin, 2010) The sample was attached to a gas sensor boat. The initial temperature was 25°C and a maximum of 300°C on a monitor screen at 100, 500, and 1,000× magnification and then photographed. Meanwhile, thermal degradation analysis was used to determine the changes in powder mass using a simultaneous TGA type STA 6000. The powdered leaf of the sengkubak was weighed at 6–7 g and then heated to a maximum temperature of 750°C with a heating rate of 10°C/min, stabilizing the powder by the change in weight with temperature changes.

2.5 Data analysis

Quantitative data were obtained through research and analyzed using ANOVA (analysis of variance/using Statistical Package for the Social Sciences [SPSS] 25), and if the results were significantly different, it was continued with the DUNCAN test at a significance level of 5%.

3 RESULTS AND DISCUSSION

3.1 Dry weight yield

Drying fresh ingredients through heating is one way to reduce the water content. The percentage of the dry weight of sengkubak leaves resulting from air drying, on average, is 20.24–21.29%, racks covered with a black cloth under sunlight is 18.75–20.80%, and open oven (natural gas) is 25.94–27.10%. The drying method had a significant effect of < 0.05 on the dry weight of sengkubak leaves. Drying in an open oven produces the highest yield. However, the dry weight of the drying results between the air dryer and the para-para covered with black cloth under sunlight was not significantly different (Table 1). These results correspond with Sinaga et al. (2024), and the percentage of drying rate of guava "klutuk" leaves is higher using the oven technique compared to air drying. Meanwhile, the dry weight percentage is based on the level of leaf maturity, namely, young leaves 20.19–25.94%, mature leaves 18.75–25.94%, and old leaves 20.80–27.10%. Old leaves produce a higher dry weight yield than young and mature ones. The results are in line with the statement of Okfrianti et al. (2022). The highest yield of 27.10% was obtained from old leaves dried using the open oven method. The dry weight between different levels of leaf maturity dried using the same drying method was not significantly different at the 5% test level. The drying method has a significant effect on the dry weight yield of pandan leaves (Purwanti et al., 2018).

3.2 Moisture content

Simplicia is a form of product processed from fresh ingredients. Drying is one method to obtain simplicia because it can reduce the water content of the material to a certain extent. Reducing the water content in fresh ingredients can prevent quality degradation. The initial moisture content of sengkubak leaves was 53.03%. After drying using the air-drying method, the average moisture content decreased to 11.94%, with sunlight covered with black cloth amounting to 9.63%, and an open oven form of 6.92%. Oven drying produces simplicia with lower water content than the other two drying methods. The drying method has a very significant effect of < 0.05 on the moisture content of sengkubak simplicia leaves as shown in Table 1. The drying results on the level of maturity of leaves are shown in Table 2; the average water content of young leaves was 9.66%, medium 9.56%, and old 9.27%. The moisture content of young and mature leaves was higher than that of old leaves. The research results are similar to Widarta and Wiadnyani's statement (2019), namely, that the moisture content of young avocado leaves is higher than old. However, based on the statistical analysis, the water content of simplicia between levels of leaf maturity was not significantly different. The level of leaf maturity did not have a significant effect (> 0.05) on water content. Based on the level of leaf maturity, the water content of sengkubak leaf simplicia is below 10% and has met the standard value.

3.3 Color

Fresh sengkubak leaves at various stages of maturity before drying are shown in Figure 1. Visual observation showed that the color of sengkubak leaves dried using different methods produced the same color of simplicia. After simplicia is processed into powder form, there are visible differences in color between drying methods. To further ensure that there were color differences between drying methods, each powder was extracted with ethanol solvent. As a result of visual observation of the color of the extract solution, air drying produced a greener color of sengkubak simplicia leaves compared to the oven method. The color of the leaves between wind-dried and covered with black cloth dried in the sun is not much different (Figure 1). Drying in the wind and sun covered with black cloth takes longer than in the oven, but the color of the simplicia and the resulting extract solution is greener. Drying temperature affects the color of simplicia sengkubak leaves.

3.4 Protein content

The results of observations on the protein content of sengkubak leaves simplicia based on the drying method, namely, air drying ranged from 20.94 to 22.49%, racks covered with a

Table 1. Effect of leaf maturity level and drying method on the yield of sengkubak leaf simplicial*.

Drying method	Quality of sengkubak leaf simplicia								
	Dry weight yield (%)			Moisture content (5)			Protein content (%)		
	Young leaf	Mature leaf	Old leaf	Young leaf	Mature leaf	Old leaf	Young leaf	Mature leaf	Old leaf
Air drying	20.35^{b}	20.24^{b}	21.29^{b}	12.00^{a}	11.94^{a}	11.88^{a}	20.94^{cd}	21.46a^{bc}	22.49^{a}
Racks covered with a black cloth sunlight	20.19^{b}	18.75^{b}	20.80^{b}	10.05^{a}	9.70^{a}	9.14^{a}	22.38^{ab}	22.45^{a}	19.91^{de}
Open oven (natural gas)	25.94^{a}	25.94^{a}	27.10^{a}	6.92^{b}	7.05^{b}	6.79^{b}	19.30^{e}	18.86^{e}	20.85^{cd}

*Numbers followed by the same letter in the same column are not significantly different at the 5% DMRT level; DMRT: Duncan's Multiple Range Test.

black cloth under sunlight ranged from 19.91 to 22.45%, and oven-open in box form (natural gas) ranged from 18.86 to 20.85%. The highest protein content was found in simplicia dried using the air-drying method, followed by the racks method covered with a black cloth under sunlight and the open-oven box method (natural gas). Based on data analysis with SPSS 25, the drying method affects protein content. Furthermore, the results were further processed using the Duncan test; the drying method had a significant effect of < 0.05 on the protein content of sengkubak leaf simplicia. Furthermore, based on the level of leaf maturity, the protein content of young leaves is around 19.30–22.38%, medium leaves 18.86–22.45%, and old leaves 19.91–22.49%. The protein content in old leaves is higher than in semi-old and young leaves. The level of leaf maturity has a significant effect of < 0.05 on protein levels, as shown in Table 1. The drying method and the level of leaf maturity influence the protein content of sengkubak leaves, but they do not interact with each other.

3.5 Antioxidant activity

The strength of the antioxidant activity of sengkubak leaf simplicia in capturing free radicals can be seen from the IC_{50} value. Drying leaves using the air-drying method obtained IC_{50} values ranging from 288.66 to 289.19 ppm, racks covered with black cloth under sunlight from 267.04 to 301.12 ppm, and open oven (natural gas) from 249.19 to 271.20 ppm. Drying leaves using the open oven (natural gas) produces a smaller IC_{50} value than the air-drying method and a rack covered with black cloth under sunlight. The results of statistical analysis using SPSS 25 show that the IC_{50} values between drying methods are significantly different. The drying method had a significant effect of < 0.05 on the antioxidant activity of sengkubak leaf simplicia, as shown in (Table 2). Furthermore, based on the level of leaf maturity, the IC_{50} value for young leaves ranges from 271.20 to 301.66 ppm, mature leaves from 249.19 to 289.04 ppm, and old leaves from 250.50 to 288.66 ppm. The results of the statistic of data analysis using SPSS 25 showed that the level of leaf maturity did not provide significant results for the IC_{50} value. The level of leaf maturity did not have a significant effect of > 0.05 on the IC_{50} value of sengkubak leaves simplicia.

3.6 Scanning electron microscopy of sengkubak leaf powder

Observations on changes in the morphological structure of sengkubak leaf powder were only carried out on simplicia, resulting from drying using the para-para method covered with black cloth (sunlight) containing optimal protein levels using a SEM tool. Display of the morphological structure of young, medium, and old sengkubak leaf powder at 1,000× magnification. The results of the photo shoot are shown in Figure 2. The morphological structure of old leaf powder is smaller than

Table 2. Effect of leaf maturity level and drying method on the antioxidant activity of sengkubak leaf simplicial*.

Drying method	Young leaves			Mature leaves			Old leaves		
	Line equation	R^2	IC_{50} value (ppm)	Line equation	R^2	IC_{50} value (ppm)	Line equation	R^2	IC_{50} value (ppm)
A	Y = 0.1633x + 2.7717	0.9946	289.19^{de}	Y = 0.1613x + 3.3779	0.9976	289.04^{de}	Y = 0.1605x + 3.67	0.9958	288.66^{de}
B	Y = 0.153x + 3.93	0.9983	301.12^{e}	Y = 0.1726x + 3.9087	0.9939	267.04^{c}	Y = 0.1604x + 5.2283	0.9959	279.15^{cd}
C	Y = 0.1664x + 4.87	0.9944	271.20^{c}	Y = 0.1819x + 4.6735	0.9970	249.19^{ab}	Y = 0.1633x + 2.7717	0.9955	250.50^{ab}

*Numbers followed by the same letter in the same column are not significantly different at the 5% DMRT level. Values followed by different letters are significantly different; A: Air drying; B: Rack covered with a black cloth under sunlight; C: Open oven; DMRT: Duncan's Multiple Range Test.

M: Young; S: Mature; T: Old.

Figure 1. Color of sengkubak leaf extract from various drying methods and leaf maturity.

that of young and medium leaves. At 1,000× magnification, the powder surface morphology of young and old sengkubak leaves is porous/hollow, while the mature leaves are stem-shaped.

3.7 Thermogravimetric analysis of sengkubak leaf powder

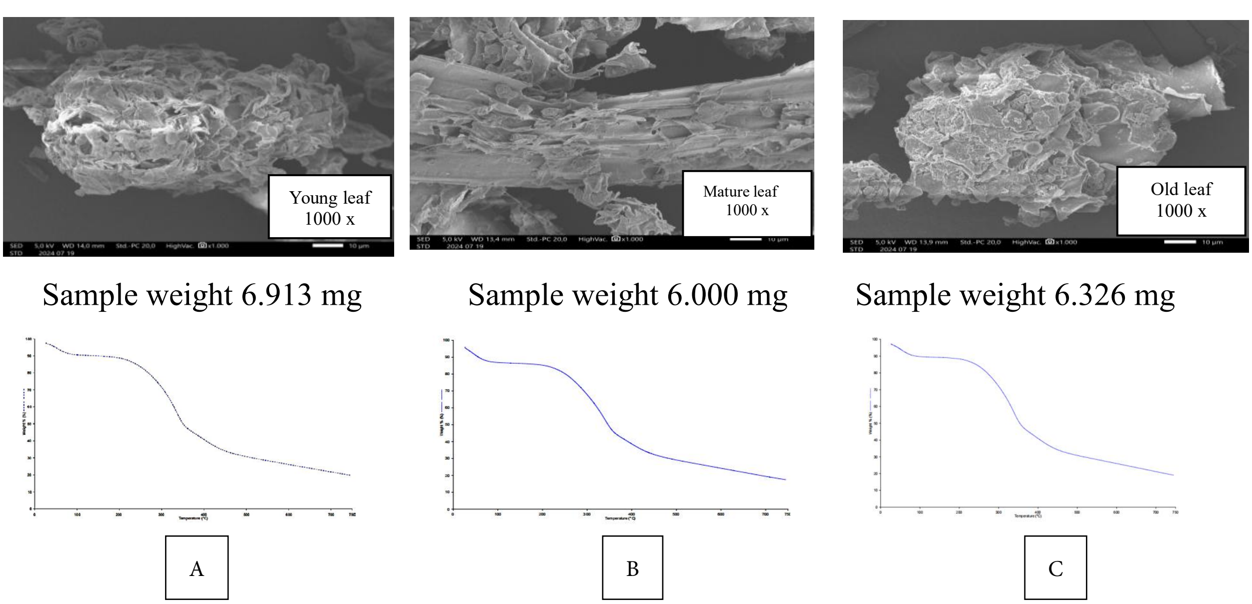
TGA analysis was carried out on leaf powder to determine the decrease in mass at various levels of leaf maturity after going through the heating process. The leaves in TGA are the result of drying on para-para covered with black cloth in the sun. The thermogram of TGA results of leaf powder from various stages of maturity is shown in Figure 2. Powder mass changes of up to 50% occurred at a temperature of 366.312,5°C with a heating time of 34.13 min, medium leaves at a temperature of 370.291,6°C for 34.52 min, and old leaves at a temperature of 364.041,6°C for 33.904 min. Some samples that are heated do not experience evaporation. Heating at high temperatures causes degradation so that the weight is reduced. The percentage of weight loss between young, medium, and old leaves was not significantly different.

3.8 Chemical compounds

Chemical components are identified to determine the chemical compounds present in dried sengkubak leaves using various drying methods so that their use becomes wider. The results of chemical component identification using the GC-MS method: the first chemical compound detected in air-dried young leaves was D-Limonene at 3.64% with a retention time (RT) of 4,638 min, the highest peak was squalane at 4.69% RT 35,009, and the highest concentration is 9,12,15-octadecatrienoic acid (Z, Z, Z) 13.99 RT 32,177. The first chemical compound in mature leaves is Neophytadiene 4.18% RT 29,135, the highest peak is the compound 4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol 5.68% RT 34,490, and the highest concentration is heptadecanoic acid, methyl ester 20.40% RT 31,171. Meanwhile, in old leaves, D-Limonene was 1.29%, RT 4,577, the highest peak, and the highest concentration was the patchouli alcohol compound, 42.25%, and the retention time was 23,197. Identification results are shown in Table 3.

The first chemical compound detected in young leaves dried on a rack covered with a black cloth under sunlight was cetene 12.93% with a retention time of 29,000 min, the highest peak and, at the same time, the highest concentration was cis-13-octadecanoic acid, 28.91% methyl ester, RT of 31.748 min. Identification results are shown in Table 4. In semi-old leaves, the first is D-Limonene 3.44% with an RT of 4,750, the highest pic peak is the compound 4-{4-(1,3-benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol 2.34% RT 34,476, and the largest concentration is 9,12,15-octadecatrienoic acid, (Z, Z, Z) 8.86% RT 32,166. Meanwhile, in old leaves, the first compound was D-Limonene 3.45% RT of 4.684, the highest peak compound was benzene propanoic acid, 3,5-bis (1,1-dimethyl ethyl)-4hydroxy-, methyl ester 7.90% RT 30,464, and the highest concentration was the compound 9,12,15-octadecatrienoic acid, (Z, Z, Z) 11.10% RT 32,169 min.

The results of the identification of young leaves of sengkubak plants, resulting from drying in an open oven (natural gas) showed that the chemical compound that appeared first was D-Limonene at 1.42 with a retention time of 4,553 min; the compound had the highest pic peak, namely, 4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol 9.24% retention time 34,490, and the compound with the highest concentration is 9,12,15-octadecatrienoic acid, (Z, Z, Z) 12.27% retention time 32,196 min. Chemical compounds in semi-old leaves: the first to appear was D-Limonene (1.44% retention time 4,750), the highest peak compound was Cinnamyl cinnamate 6.07%



A: Young; B: Mature; C: Old.

Figure 2. SEM of sengkubak leaf powder at a magnification of 1,000× and thermal gravimetric analysis from 25 to 750°C at 10°C/min from various leaf maturity levels.

Table 3. Chemical compounds identified in sengkubak leaves using the GC-MS method as a result of air drying.

Young leaves			Mature leaves			Old leaves		
Chemical compounds	**%**	**RT**	**Chemical compounds**	**%**	**RT**	**Chemical compounds**	**%**	**RT**
Squalane	4.69	35.009	4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2 methoxyphenol	5.68	34.490	Patchouli alcohol	42.25	23.197
Benzene propanoic acid, 3,5-bis(1,1-dimethylethyl)-4-hydroxy-, methyl ester	7.17	30.468	Heptadecanoic acid, methyl ester	20.40	31.171	Phytol	2.75	31.779
Phytol	5.13	31.780	9,12,15-octadecatrienoic acid, (Z,Z,Z)	10.25	32.194	Benzene propanoic acid, 3,5-bis (1,1-dimethylethyl)-4 hydroxy-, methyl ester	4.51	30.467
9,12,15-octadecatrienoic acid, (Z,Z,Z)	13.09	32.177	Neophytadiene	4.18	29.135	Neophytadiene	5.33	29.132
Vit E	7.27	36.972	Cholesterol	4.28	36.935	Squalene	2.05	35.005
Neophytadiene	6.60	29.128	Phytol	2.36	31.784	Vitamin E	4.73	36.973
Stigmasterol	5.62	38.138	hexadecanoic acid, methyl ester	3.59	30.292	4-{4-(1,3-Benzodioxol-5-y)-2,3-dimethylbuthyl}-2-methoxyphenol	1.82	34.472
D-limonene	3.64	4.638	Benzene propanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	4.27	30.472	n-Hexadecanoic acid	2.34	30.955
Gamma terpinene	2.45	5.039	Vitamin E	2.61	36.968	9,12-Octadecatrienoic acid (Z,Z)	4.94	32.141
Gamma sitosterol	3.39	38.720	Stigmasterol	2.40	38.133	D-limonene	1.29	4.577
Eicosane	2.32	36.499	Lupeol	1.52	39.622	Gamma-terpinene	1.53	4.984
Lupeol	2.38	39.633				Stigmasterol	2.16	38.128

RT: retention time.

Table 4. Chemical compounds identified in sengkubak leaves using the GC-MS method as a result of drying on a rack covered with a black cloth under sunlight.

Young leaves			Mature leaves			Old leaves		
Chemical compounds	**%**	**RT**	**Chemical compounds**	**%**	**RT**	**Chemical compounds**	**%**	**RT**
Cis-13-Octadecanoic acid, methyl ester	28.91	31.748	4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol	2.34	34.476	Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	7.90	30.464
Hexadecanoic acid, methyl ester	19.78	30.362	Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	5.28	30.468	4-{4-(1,3-Benzodioxol-5-y)-2,3-dimethylbuthyl}-2-methoxyphenol	4.97	34.476
Methyl stearate	4.73	31.866	Neophytadiene	7.19	29.130	Neophytadiene	7.80	29.123
Cetene	12.93	29.900	3-Methyldodecanoic acid	3.58	30.973	9,12,15-Octadecatrienoic acid, (Z,Z,Z)	11.10	32.169
Neophytadiene	1.98	29.192	9,12,15-Octadecatrienoic acid, (Z,Z,Z)	8.86	32.166	3-Methyldodecanoic acid	4.91	30.965
Phytol	1.19	31.796	Squalene	1.80	35.006	Phytol	2.81	31.778
Vitamin E	1.04	36.962	Vitamin E	3.60	36.968	Gamma-terpinene	2.93	5.081
Hexadecanoic acid, 15-methyl-methyl ester	7.74	31.189	D-Limonene	3.44	4.750	Vitamin E	3.90	36.962
9-Hexadecanoic acid, methylester (Z)	3.44	30.079	Gamma-Terpinene	1.71	5.141	Squalene	1.58	35.004
Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	1.83	30.494	Stigmasterol	3.72	38.133	Eucalyptus	1.28	4.724
			Octadecatrienoic acid	1.50	32.247	D-Limonene	3.45	4.684
			Gamma sitosterol	2.22	38.720	Stigmasterol	3.15	38.125
						Octadecanoic acid	1.90	32.245
						9,12,15-Octadecatrienoic acid, (Z,Z,Z)	1.63	31.674
						Lupeol	2.74	39.622
						Gamma sitosterol	2.11	38.712
						Tetradecanoic acid	4.23	28.519

RT 33,417, and the highest concentration was the compound 9,12,15-Octadecatrienoic acid, (Z, Z, Z) 9.47% retention time 32,192 min. Identification results are shown in Table 5. Meanwhile, in old leaves, the first compound to appear was phenol, 2,6-dimethoxy-4-(2-prophenyl) 1.51% retention time 22,109 and the compound had the highest pic peak, and the largest concentration, namely, 9,12-octadecatrienoic acid (Z, Z) 33.76% with a retention time of 32,435 min. Identification results are shown in Tables 3–5.

3.9 Discussion

3.9.1 Weight yield

To maintain the quality of fresh ingredients, it can be done by reducing the water content of the ingredients. One of the processes of reducing water content can be done through the drying process. Apart from the water content, the weight of the material and the chemical compounds contained in the material are also reduced due to heat. The drying method is related to temperature and humidity and influences the drying speed. The yield of the dry weight of simplicia sengkubak leaves from drying in an open oven with a heat source from natural gas is higher than the wind and sun drying method covered with black cloth. The average temperature of the drying method during the drying process is air dry 30.7°C, black cloth sunlight collector 42.1°C, and open oven 47.5°C. The higher the temperature, the faster the drying process, and the lower the temperature, the longer the drying process takes place so that the weight of the material decreases. Producing sengkubak leaf simplicia using an oven dryer takes around 9 h, 16 h in the sun covered with black cloth, and 43 h in the wind. According to Winangsih et al. (2013), oven drying can produce constant dry weight more quickly. The percentage of dry weight yield using the para-para method covered with black cloth (sunlight) is smaller than in the oven method (Wiraguna et al., 2015).

The dry weight yield percentage of old sengkubak leaves was higher than that of young and medium leaves, but in the

Table 5. The chemical compounds identified in sengkubak leaves using the GC-MS method as a result of drying using an open oven (natural gas).

Young leaves			Mature leaves			Old leaves		
Chemical Compound	**%**	**RT**	**Chemical Compound**	**%**	**RT**	**Chemical Compound**	**%**	**RT**
4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol	9.24	34.490	Cinnamyl cinnamate	6.07	33.417	9,12-Octadecatrienoic acid (Z,Z)	33.76	32.435
9,12,15-Octadecatrienoic acid, (Z,Z,Z)	12.27	32.196	Phytol	8.40	31.785	11-Octadecanoic acid, methyl ester	11.88	31.729
Phytol	3.30	31.783	4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2 methoxyphenol	4.09	34.482	4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2 methoxyphenol	3.24	34.512
Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	4.87	30.469	Neophytadiene	7.48	29.136	(1S,2R)-2-(4-Allyl-2,6-dimethoxyphenoxy)-1-(3,4-dimethoxyphenyl) propyl acetate	3.44	35.229
N-Hexadecanoic acid-	11.18	30.981	9,12,15-Octadecatrienoic acid, (Z,Z,Z)	9.47	32.192	Hexadecanoic acid, methyl ester	2.51	30.320
Gamma terpinene	1.96	4.961	Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4hydroxy-, methyl ester	5.17	30.470	Tetradecanoic acid	7.19	29.150
D-limonene	1.42	4.553	Hexadecanoic acid, methyl ester	6.00	30.294	4-{4-(1,3-Benzodioxol-5-y)-2,3-dimethylbuthyl}-2-methoxyphenol		
9,12-Octadecatrienoic acid (Z,Z)	2.65	31.631	Vitamin E	4.99	36.975	n-Hexadecanoic acid	2.34	30.955
Vitamin E	2.72	36.962	(1S,2R)-2-(4-Allyl-2,6-dimethoxyphenoxy)-1-(3,4-dimethoxyphenyl) propyl acetate	2.60	35.191	N-Hexadecanoic acid	11.88	31.133
Stigmasterol	2.73	38.130	Stigmasterol	4.76	38.145	Vitamin E	1.83	36.995
Tetradecanoic acid	8.84	28.762	Cis-13-Octadecanoic acid, methyl ester	1.92	31.674	(1S,2R)-2-(4-Allyl-2,6-dimethoxyphenoxy)-1-(3,4-dimethoxyphenyl) propyl acetate	1.16	35.896
			Squalene	1.52	35.006	N-(4-Methoxyphenyl)-2 hydroxymino-acetamide	1.38	35.320
			D-Limonene	1.44	4.750	Benzenepropanoic acid, 3,5-bis (1,1-dimethylethyl)-4 hydroxy-, methyl ester	1.07	30.493
			Gamma terpinene	1.66	5.142	Phenol,2,6-dimethoxy-4-(2-prophenyl)	1.51	22.109

RT: retention time.

statistical analysis, the dry weight yield was not significantly different between levels of leaf maturity. According to Whiteman, P. C. (1974) in Sylvia et al. (2021), plant dry matter production is influenced by nitrogen levels. The higher the nitrogen content of the dried material, the higher the resulting dry weight. According to Septyaningsih et al. (2014), young leaves contain low fiber and high water content, while old leaves, according to Savitri et al. (2017), contain a smaller moisture content and a higher proportion of cell walls compared to the cell contents. The higher the cell wall content of a plant, the more dry matter it contains.

3.9.2 Moisture content

The drying process by an oven method is the fastest to produce simplicia because the heat is more stable, whereas other dryers have fluctuating heat and depend on the weather, so the process takes a long time. Drying brown leaves at different temperatures produces simplicia with different water content (Rusyananti, 2018). Drying at high temperatures can speed up the process of water evaporation so that the material dries quickly and has a low water content (Winangsih et al., 2013). The moisture content of simplicia according to the standard is a maximum of 10%. The moisture content of leaf simplicia results from drying using an open oven and sunlight covered with black cloth meets Indonesia National Standard (SNI) standards, namely, a maximum of 10% (Departemen Kesehatan Republik Indonesia, 2017). The speed of the drying process is influenced by temperature and humidity. The dryer method related to temperature gives an effect of 93.1% and humidity 75.3%. Air at high temperatures takes up more water quickly from the material being dried so that the drying process is faster and the water content is lower. Temperature affects the drying speed; the higher the temperature, the faster the process of water evaporation from the surface of the material so that the drying process is faster than at low temperatures. In air drying, the average temperature of the dryer to produce dry sengkubak leaves is around 30.7°C, on top of a roof covered with a black cloth under sunlight is 42.1°C, and an open oven (natural gas) is 47.5°C. Meanwhile, the drying time in sequence is 43, 16, and 9 h. The simplicia is obtained from the drying process to a certain water content. In making simplicia, the quality indicated by the chemical compounds in the important ingredients is maintained, except for the water content. The young Moringa leaf powder contains a higher water content than the old (Agamou et al., 2015).

3.9.3 Color

Temperature and drying time influence the color of simplicia. According to Purwanti et al.(2018) the longer the drying process, the longer water remains in the simplicia, and the greater the possibility of chlorophyll degrading into pheophytin. To ensure differences in powder color between drying, extraction was performed. Drying temperature affects the color of sengkubak simplicia leaves. The drying process using the air-drying method takes around 43 h, while the racks method was used for extraction. Drying temperature affects the color of sengkubak simplicia leaves. The drying process using the air-drying method takes around 43 h, while the para-para method was used for extraction. In both drying methods, each powder was extracted. Drying temperature affects the color of sengkubak simplicia leaves. The drying process using the air-drying method takes around 43 h, the racks method covered with a black cloth under sunlight takes 16 h, and the open-oven method (natural gas) takes 9 h. Meanwhile, the average temperature of the drying method during the drying process is 30.7, 42.1, and 47.5°C. Visual observation shows that the color of the solution resulting from the extraction of sengkubak leaves powder differs between drying methods (Figure 1). The results show that the color of simplicia differs between drying methods. Drying using the air-dryer method and shelves covered with black cloth under sunlight produces an extract that is greener in color than in an open oven (natural gas). Drying temperature affects the color of sengkubak simplicia leaves. Meanwhile, the color of extracts from different levels of leaf maturity is the same, except for the results of drying in an open oven (natural gas). Temperature and light greatly influence chlorophyll stability. This statement is in line with the result of research, where the open oven drying method (natural gas) has a higher temperature than air drying, and the racks covered with black cloth. Drying at high temperatures can cause chlorophyll degradation to pheophytin and affect the color of the dried product (Ali et al., 2014; Purwanti et al., 2018). Temperature has a significant effect on color (Zhang et al., 2020).

3.9.4 Protein content

Protein levels are related to amino acids, one of which is glutamic acid (Murti et al., 2021). Glutamic acid levels can decrease due to heating at high temperatures for a long time because protein denaturation occurs (Viyanti et al., 2019). Heat accelerates the pheophytination reaction because heat can denature proteins. The average temperature of the drying method during the drying process is 30.7°C for air drying, 42.5°C for racks covered with a black cloth under sunlight, and 47.5°C for the open-oven method. According to Li et al. (2014), protein levels are related to nitrogen levels and can be during the heating process. The higher the drying temperature, the nitrogen content of simplicia decreases because the color changes from green to brownish. The changes in leaf color due to heating can reduce nitrogen and protein levels due to protein denaturation. Apart from that, the color of the leaves (fresh material) before drying also influences the protein content after drying (simplicia) (Novia et al., 2011). The color of fresh plant leaves can vary depending on the level of maturity and is related to chlorophyll. The young leaves of the sengkubak plant are light green, the older leaves are green, and the older leaves are slightly yellowish green, as shown in Figure 3. The results of the research are commensurate with the statement of Saputri et al. (2019), namely, that the protein content of old leaves of Moringa plants is higher than in young leaves. The optimal drying method for sengkubak leaves to produce simplicia with maximum protein content is that young or mature leaves are dried on racks under sunlight and covered with a black cloth.

3.9.5 Antioxidant activity

According to Widarta and Wiadnyani (2019), the level of leaf age and drying method affects the bioactive compounds and antioxidant activity contained in avocado leaf extract. Long drying at room temperature can reduce the levels of simplicia bioactive compounds (Winangsih et al., 2013). According to Bernard et al. (2014), dry old leaves have stronger antioxidant activity to capture free radicals than young leaves. Phenolic acids can accumulate at each stage of leaf development so that the total phenolic content increases, which has different effects on the antioxidant activity of certain plant extracts (Chang et al., 2018). The IC_{50} value is inversely proportional to antioxidant activity. The smaller the IC_{50} value, the stronger the antioxidant activity. The strongest antioxidant activity is found in mature leaves that are dried in an open oven (natural gas) or on racks covered with a black cloth under sunlight. The smaller the IC_{50} value of simplicia, the stronger its antioxidant activity. The IC_{50} value is inversely proportional to antioxidant activity. The IC_{50} value is inversely proportional to antioxidant activity. The smaller the IC_{50} value, the stronger the antioxidant activity.

3.9.6 Chemical component

Chemical compounds contained in plants have diverse biological properties. Chemical compounds contained in plants have different pharmacological effects (Victor & David, 2015). According to Wu et al. (2019), plant secondary metabolites can play a role in protecting themselves from pathogen attacks. Patchouli alcohol compounds belong to the sesquiterpene group. They are useful as a fixative and anti-viral (Isnaini et al., 2022; Lu et al., 2016), anti-depressant (Zhuo et al., 2020), and anti-inflammatory (Zhang et al., 2020). The concentration of 9,12,15-octadecatrienoic acid, (Z, Z, Z) in young leaves ranges between 12.27 and 13.00%, and in mature, old 8.86 and 10.25%, has anti-inflammatory, anti-bacterial, anti-cancer, hepatoprotective and antioxidant properties (Amala & Jeyaraj, 2014; Rani & Kapoor, 2019). The largest average concentration was found in mature leaves. The chemical compound hexadecanoic acid methyl ester is anti-bacterial, and octadecanoic acid is anti-fungal (Shaaban et al., 2021). The chemical compound neophytadiene functions as an anti-pyretic, analgesic, anti-inflammatory, anti-microbial, and antioxidant (Anjali et al., 2019; Bhardwaj et al., 2020; El Shafay et al., 2015). The concentration ranges from 1.98 to 6.60% in young leaves, 4.18 to 7.48% in mature leaves, and 4.38 to 7.80% in old leaves. Phytol is around 1.19–5.13% in young leaves, 2.36–8.40 in mature leaves, and 2.75–2.81 in old leaves. According to Ogunlesi et al. (2009),the phytol compound is useful as well as an antioxidant and preventative and therapeutic for arthritis. The chemical compound phytol can also function as an anti-diabetic (Goto et al., 2010). The D-Limonene compound can used as a food flavoring, has a citrus-like aroma, and can be applied in the chemical industry, cosmetics, and cleaning products (Ruiz & Flotats, 2014; Ruiz et al., 2016). In the Code of Federal Regulations, the compound D-Limonene is recognized as safe (generally recognized as safe [GRAS]) as a flavoring agent. In addition, D-limonene is useful as an antioxidant (Toscano-Garibay et al., 2017), anti-cancer (prostate cancer) (Ye et al., 2020), antidiabetic (Bacanlı et al., 2017), anti-gastroprotective (Vieira et al., 2018). D-limonene concentrations ranged from 3.44 to 3.64%. The level of leaf maturity did not affect the D-limonene concentration, but the drying method did. Vitamin E has stronger antioxidant activity than β-carotene (Xu et al., 2009). Young and mature leaves that result from air drying contain greater concentrations of vitamin E than old leaves and other drying methods. The compound 4-{4-(1,3-Benzodioxol-5-yl}-2,3-dimethylbutyl}-2methoxyphenol is used as an antioxidant and flavor (Suryanti and Anwar, 2008). The largest concentration is there in mature leaves dried in an open oven, namely, 4.04%. The compound Benzenepropanoic acid, 3,5-bis(1,1-dimethyl ethyl)-4-hydroxy-, methyl ester has biological properties as an anti-microbial (Błaszczyk et al., 2021). The concentration was in mature (4.90%) and young (4.62%) leaves resulting from the air-drying method. Meanwhile, the Cinnamyl cinnamate compound is an antioxidant, anti-cancer, and cardiovascular agent. 9,12 Octadecanoic acid (Z, Z), methyl ester is used as an anti-inflammatory and arthritis agent (Singh & Chaturvedi, 2019). The largest concentration, 2.65%, was found in young leaves dried in an open oven. Stigmasterol originates from sitosterol (Aboobucker & Suza, 2019) and has antioxidant, hypoglycemic, thyroid inhibitor, progesterone precursor, anti-microbial, anti-cancer, anti-rheumatism, anti-asthama, anti-inflammatory, and diuretic properties (Tyagi & Agarwal, 2017). The largest concentration, on average, 3.63%, was found in mature leaves resulting from air drying. Exploration of active ingredients from plants is generally safer than chemicals because they have fewer side effects (Ma'ruf et al., 2018). Herbal-based medicines: their use is supported by WHO for health purposes (Sudirman & Skripsa, 2020).

3.9.7 The scanning electron microscopy and thermogravimetric analysis sengkubak leaves powder

TGA was carried out to determine the effect of heat on changes in material mass. The higher the heating temperature, the higher the percentage of mass change in the material. Reducing the mass of the material by up to 50% requires temperature and heating time that are not significantly different between young, medium, and old leaves. According to Sudarno (2014), changes in mass can occur due to the decomposition of heated material consisting of devolatilization, oxidation, or reduction depending on the surrounding atmospheric environment. The faster and more mass loss indicates that the material is more reactive (Zhao et al., 2011). Several factors that influence the TGA measurement results are heating rate, number/mass of samples, particle size, type, and flow rate of the atmosphere (gas). Thermogravimetric analysis is very important to determine the appropriate drying temperature limits.

4 CONCLUSION

The drying method influences the yield of dry weight, water content, simplicia color, protein content, antioxidant activity, chemical components, temperature on the morphological character, and mass changes of the material. The quality of simplicia between levels of leaf maturity was not significantly different. Sengkubak leaves that are aired or dried in the sun and covered with black cloth can produce quality leaf simplicia. The color of young leaf extract is greener than that of old leaves using the open oven method. The average dry weight yield of leaves ranged from 21.16 to 23.06%, water content was 9.27–9.66%, IC50 value was 268.42–287.17 ppm, and protein was 20.87–21.08% in young and mature leaves. The number of chemical components detected in young and medium leaves was greater than in old leaves. Based on the identification results, sengkubak leaves contain the chemical compounds D-Limonene, neophytadiene, phytol, benzene-propanoic acid, 3,5-bis(1,1,-dimethyl ethyl)-4-hydroxy-, methyl ester, 9,12,15 octadecatrienoic acid, hexadecanoic acid, methyl ester, vitamin E, 9,12 octadecanoic acid (Z, Z), stigmasterol, gamma-terpinene, gamma stigmasterol, squalene, and 4-{4-(1,3- Benzidioxol-5-yl)-2,3-dimethyl butyl}-2 methoxy phenol.

REFERENCES

Aboobucker, S. I., & Suza, W. P. (2019). Why do plants convert sitosterol to stigmasterol? *Frontiers in Plant Science*, *10*, 354. https://doi.org/10.3389/fpls.2019.00354

Agamou, J. A. A., Fombang, E. N., & Mbofung, C. M. (2015). Particular benefits can be attributed to *Moringa oleifera* Lam leaves based on origin and stage of maturity. *Journal of Experimental Biology and Agricultural Sciences*, *3*(6), 541-555. https://doi.org/10.18006/2015.3(6).541.555

Ali, M. A., Yusof, Y. A., Chin, N. L., Ibrahim, M. N., & Basra, S. M. A. (2014). Drying kinetics and colour analysis of *Moringa oleifera* leaves. *Agriculture and Agricultural Science Procedia*, *2*, 394-400. https://doi.org/10.1016/j.aaspro.2014.11.055

Amala, E. U., & Jeyaraj, M. (2014). Comparative evaluation of phytocomponents present in the methanolic extract of *Terminalia chebula* Retz., *Terminalia bellirica* Roxb., and *Phyllanthus emblica* L., fruit extracts using GC-MS analysis. *International Journal of Pharma and Bio Sciences*, *5*(4), 927-934.

Anjali, K. P., Sangeetha, B. M., Devi, G., Raghunathan, R., & Dutta, S. (2019). Bioprospecting of seaweeds (*Ulva lactuca* and *Stoechospermum marginatum*): The compound characterization and functional applications in medicine—a comparative study. *Journal of Photochemistry & Photobiology B: Biology*, *200*, 111622. https://doi.org/10.1016/j.jphotobiol.2019.111622

Bacanlı, M., Anlar, H. G., Aydın, S., Çal, T., Arı, N., Ündeğer Bucurgat, Ü., Başaran, A. A., & Başaran, N. (2017). d-Limonene ameliorates diabetes and its complications in streptozotocin-induced diabetic rats. *Food and Chemical Toxicology*, *110*, 434-442. https://doi.org/10.1016/j.fct.2017.09.020

Bernard, D., Kwabena, A. I., Osei, O. D., Daniel, G. A., Elom, S. A., & Sandra, A. (2014). The effect of different drying methods on the phytochemicals and radical scavenging activity of Ceylon Cinnamon (*Cinnamomum zeylanicum*) plant parts. *European Journal of Medicinal Plants*, *4*(11), 1324-1335. https://doi.org/10.9734/EJMP/2014/11990

Bhardwaj, M., Sali, V. K., & Mani, S. (2020). Neophytadiene from *Turbinaria ornata* suppresses LPS-induced inflammatory response in RAW 264.7 macrophages and Sprague Dawley rats. *Inflammation*, *43*(3), 937-950. https://doi.org/10.1007/s10753-020-01179-z

Błaszczyk, N., Rosiak, A., & Kałużna-Czaplińska, J. (2021). The potential role of cinnamon in human health. *Forests*, *12*(5), 648. https://doi.org/10.3390/f12050648

Boldin, P. (2010). *Electric field measurements using scanning electron microscope* (Master's thesis, Faculty of Technology, Lappeenranta University of Technology).

Chang, X., Lu, Y., Lin, Z., Qiu, J., Guo, X., Pan, J., & Abbasi, A. M. (2018). Impact of leaf development stages on polyphenolics profile and antioxidant activity in *Clausena lansium* (Lour.) Skeels. *BioMed Research International*. https://doi.org/10.1155/2018/7093691

Departemen Kesehatan Republik Indonesia (2017). *Farmakope Herbal Indonesia Edisi Kedua*. Ditjen POM RI

El Shafay, S. M., Ali, S. S., & El-Sheekh, M. M. (2015). Antimicrobial activity of some seaweed species from the Red Sea, against multidrug-resistant bacteria. *Egyptian Journal of Aquatic Research*, *42*(1), 65-74. https://doi.org/10.1016/j.ejar.2015.11.006

Fahmi, A., Devianti, & Agustina, R. (2019). Study of copra and coconut oil quality in the drying process with variations in energy sources. *Unsyiah Agricultural Student Scientific Journal*, *4*(3), 95-104. https://doi.org/10.17969/jimfp.v4i3.10046

Goto, T., Takahashi, N., Hirai, S., & Kawada, T. (2010). Various terpenoids derived from herbal and dietary plants function as PPAR modulators and regulate carbohydrate and lipid metabolism. *PPAR Research*, 483958. https://doi.org/10.1155/2010/483958

Isnaini, N., Khairan, K., Faradhilla, M., Sufriadi, E., Ginting, B., Prajaputra, V., Erwan, F., Lufika, R. D., & Muhammad, S. (2022). Evaluation of physical quality of patchouli oil (*Pogostemon cablin* Benth.) body butter formulation. *Journal of Patchouli and Essential Oil Products*, *1*(1), 22-26. https://doi.org/10.24815/jpeop.v1i1.23762

Juita, N., Lovadi, I., & Linda, R. (2015). Pemanfaatan tumbuhan sebagai penyedap rasa alami pada masyarakat suku Dayak Jangkang Tanjung dan Melayu di Kabupaten Sanggau. *Protobiont*, *4*(3). https://doi.org/10.26418/protobiont.v4i3.13315

Li, N., Lu, X., Pei, H., & Qiao, X. (2014). Effect of freezing pretreatment on the processing time and quality of black garlic. *Journal of Food Process Engineering*, *38*(4), 329-335. https://doi.org/10.1111/jfpe.12156

Lu, L., Yang, C., Lu, Z., Xu, H., Qiu, J., Wu, J., Wang, J., Tong, Y., Zhu, J., & Shen, J. (2016). Molecular role of EGFR-MAPK pathway in patchouli alcohol-induced apoptosis and cell cycle arrest on A549 cells in vitro and in vivo. *BioMed Research International*, 4567580. https://doi.org/10.1155/2016/4567580

Ma'ruf, M., Mawaddah, G. A., Eriana, N. N. A., Swari, F. I., Aslamiyah, S., & Lutpiatina, L. (2018). Kelulut bee honey (*Trigona spp.*) in its activity against resistant *Staphylococcus aureus* bacteria. *Journal of Health Scales*, *9*(1).

Masriani, M., & Sunarti, J. (2019). The cytotoxic activities of the extracts of sengkubak (*Pycnarrhena cauliflora*) as apoptosis inducers to HeLa cervical cancer cells. *Journal of Chemical Natural Resources*, *1*(2). https://doi.org/10.32734/jcnar.v1i2.1256

Murti, R. W., Sumardianto, & Purnamayati, L. (2021). Effect of different salt concentrations on the glutamic acid of rebon shrimp paste (*Acetes sp.*). *Jurnal Pengolahan Hasil Perikanan Indonesia*, *24*(1), 50-59. https://doi.org/10.17844/jphpi.v24i1.33201

Novia, D., Melia, S., & Ayuza, N. Z. (2011). Study of oven temperature on protein content and organoleptic value of salted eggs. *Animal Husbandry Journal*, *8*(2), 70-76.

Ogunlesi, M., Okiei, W., Ofor, E., & Osibote, A. E. (2009). Analysis of the essential oil from the dried leaves of *Euphorbia hirta* Linn (*Euphorbiaceae*), a potential medication for asthma. *African Journal of Biotechnology*, *8*, 7042-7050.

Okfrianti, Y., Irnameria, D., & Bertalina, B. (2022). Antioxidant activity of sungkai leaves (*Peronema canescens* Jack). *Journal of Health*, *13*(2), 1-8.

Pamuji, R. W. (2015). Uji toksisitas akut extrato etanol de folha de Sengkubak (*Pycnarrhena cauliflora* Diels) contra ratas fêmeas Wistar usando método OECD 425. *Jurnal Mahasiswa Farmasi Fakultas Kedokteran UNTAN*, *3*(1).

Purba, D. M., Wibowo, M. A., & Ardiningsih, P. (2014). Aktivitas antioksidan dan sitotoksik ekstrak metanol daun sengkubak (*Pycnarrhena cauliflora* Diels). *Jurnal Kimia Khatulistiwa*, *3*(2).

Purwanti, N. U., Luliana, S., & Sari, N. (2018). The effect of drying pandan leaf simplicia (*Pandanus amaryllifolius*) on the activity of DPPH free radical scavenger (2,2-Diphenyl-1-Picrylhydrazyl). *Pharmacy Medical Journal*, *13*(2), 63-72. https://doi.org/10.26630/jk.v13i2.3200

Puspita, D., & Wulandari, T. S. (2020). Analisis senyawa bioaktif pada daun kemangi imbo (*Pycnarrhena cauliflora*) yang digunakan sebagai penyedap alami. *Jurnal Teknologi Pangan dan Gizi*, *19*(1), 35-43.

Rani, J., & Kapoor, M. (2019). Gas chromatography-mass spectrometric analysis and identification of bioactive constituents of *Catharanthus roseus* and its antioxidant activity. *Asian Journal of Pharmaceutical and Clinical Research*, *12*(3). https://doi.org/10.22159/ajpcr.2019.v12i3.30865

Ruiz, B., & Flotats, X. (2014). Citrus essential oils and their influence on the anaerobic digestion process: An overview. *Waste Management*, *34*(11), 2063-2079. https://doi.org/10.1016/j.wasman.2014.06.026

Ruiz, B., de Benito, A., Rivera, J. D., & Flotats, X. (2016). Assessment of different pre-treatment methods for the removal of limonene in citrus waste and their effect on methane potential and methane production rate. *Waste Management & Research*, *34*(12), 1249-1257. https://doi.org/10.1177/0734242x16661053

Rusyananti, Y. (2018). The effect of temperature and drying time on the quality of cocoa leaf green tea (Thesis, Faculty of Food Technology and Agroindustry, Mataram University).

Saputri, G. R., Tutik, & Permatasari, A. I. (2019). Determination of protein content in young Moringa leaves and old Moringa leaves (*Moringa oleifera* L.) using the Kjeldahl method. *Journal of Pharmaceutical Analysis*, *4*(2), 108-116.

Savitri, I., Suhendra, L., & Wartini, N. M. (2017). The effect of solvent type on the maceration method on the characteristics of *Sargassum polycystum* extract. *Journal of Agro-Industrial Engineering and Management*, *5*(3), 93-101.

Septyaningsih, E., Ardli, E. R., & Widyastuti, A. (2014). Morphometric study and herbivory level of mangrove leaves in Segara Anakan Cilacap. *Scripta Biologica*, *1*(2), 137-140. https://doi.org/10.20884/1.sb.2014.1.2.438

Shaaban, M. T., Ghaly, M. F., & Fahmi, S. M. (2021). Antibacterial activities of hexadecanoic acid methyl ester and green-synthesized silver nanoparticles against multidrug-resistant bacteria. *Journal of Basic Microbiology*, *61*(6), 557-568. https://doi.org/10.1002/jobm.202100061

Sinaga, B., Sondak, E. S., & Ningsih, A. W. (2024). Pengaruh metode pengeringan terhadap kualitas simplisia daun jambu biji merah (*Psidium guajava* L.). *Prosiding Seminar Nasional Polkesta*, *1*(1), 111-120.

Singh, R., & Chaturvedi, P. (2019). Phytochemical characterization of rhizome, fruit, leaf, and callus of *Rheum emodi* Wall. using GC-MS. *Pharmacognosy Journal*, *11*(3), 617-623. https://doi.org/10.5530/pj.2019.11.99

Sudarno (2014). *Characterization of sawdust and its mixtures under isothermal and non-isothermal conditions* [Postgraduate, Mechanical Engineering, Semarang State University].

Sudirman, & Skripsa, T. H. (2020). Utilization of traditional medicine services (Batra) as a role model for back to nature medicine in the future. *Journal of Research Applications to Society*, *1*(1), 45-50. https://doi.org/10.55583/arsy.v1i1.44

Sylvia, D., Apriliana, V., & Rasydy, L. O. A. (2021). Analysis of the protein content in guava leaves (*Psidium guajava* L.) using the Kjeldahl method and UV-VIS spectrophotometry. *Farmagazine Journal*, *8*(2), 64-72.

Toscano-Garibay, J. D., Arriaga-Alba, M., Sánchez-Navarrete, J., Mendoza-García, M., Flores-Estrada, J. J., Moreno-Eutimio, M. A., Espinosa-Aguirre, J. J., González-Ávila, M., & Ruiz-Pérez, N. J. (2017). Antimutagenic and antioxidant activity of the essential oils of *Citrus sinensus* and *Citrus latifolia*. *Scientific Reports*, *7*(1), 11479. https://doi.org/10.1038/s41598-017-11818-5

Tritisari, A., Rozana, & Maslan. (2023). Quality characteristics and organoleptic analysis of natural powder flavorings sengkubak leaves (*Pycnarrhena cauliflora* Diels). *Proceedings of the National Seminar*, *5*(1), 131-138.

Tyagi, T., & Agarwal, M. (2017). Phytochemical screening and GC-MS analysis of bioactive constituents in the ethanolic extract of *Pistia stratiotes* L. and *Eichhornia crassipes* (Mart.) Solms. *Journal of Pharmacognosy and Phytochemistry*, *6*(1), 195-206.

Victor, O. O., & David, S. M. (2015). GC-MS analysis of phyto-components from the leaves of *Senna alata* L. *Journal of Plant Science*, *3*(3), 133-136. https://doi.org/10.11648/j.jps.20150303.14

Vieira, A. J., Beserra, F. P., Souza, M. C., Totti, B. M., & Rozza, A. L. (2018). Limonene: Aroma of innovation in health and disease. *Chemico-Biological Interactions*, *283*, 97-106. https://doi.org/10.1016/j.cbi.2018.02.007

Viyanti, R., Sumardianto, & Suharto, S. (2019). The use of fish cooking water varies with the glutamic acid content in petis. *PENA Aquatics*, *18*(2), 23-33.

Widarta, I. W. R., & Wiadnyani, A. A. I. S. (2019). Pengaruh metode pengeringan terhadap aktivitas antioksidan daun alpukat. *Jurnal Aplikasi Teknologi Pangan*, *8*(3), 80-85. https://doi.org/10.17728/jatp.3361

Winangsih, W., Prihastanti, E., & Parman, S. (2013). Pengaruh metode pengeringan terhadap kualitas simplisia lempuyang wangi (*Zingiber aromaticum* L.). *Anatomi Fisiologi*, *21*(1), 19-25. https://doi.org/10.14710/baf.v21i1.6268

Wiraguna, I. G. N. P., Wartini, N. M., & Yoga, I. W. G. S. (2015). The effect of curing method and length on the characteristics of pandan wangi leaves (*Pandanus amaryllifolius* Roxb.). *Journal of Agroindustrial Engineering and Management*, *3*(2), 109-119.

Wu, S. C., Liu, F., Zhu, K., & Shen, J. Z. (2019). Natural products that target virulence factors in antibiotic-resistant *Staphylococcus aureus*. *Journal of Agricultural and Food Chemistry*, *67*(48), 13195-13211. https://doi.org/10.1021/acs.jafc.9b05595

Xu, L., Davis, T. A., & Porter, N. A. (2009). Rate constants for peroxidation of polyunsaturated fatty acids and sterols in solution and in liposomes. *Journal of the American Chemical Society*, *131*(36), 13037-13044. https://doi.org/10.1021/ja9029076

Ye, Z., Liang, Z., Mi, Q., & Guo, Y. (2020). Limonene terpenoid obstructs human bladder cancer cell (T24 cell line) growth by inducing cellular apoptosis, caspase activation, G2/M phase cell cycle arrest, and stops cancer metastasis. *Journal of BU ON*, *25*(1), 280-285.

Zhang, G., Liu, M., Song, M., Wang, J., Cai, J., Lin, C., Li, Y., Jin, X., Shen, C., Chen, Z., Cai, D., Gao, Y., Zhu, C., Lin, C., & Liu, C. (2020). Patchouli alcohol activates PXR and suppresses the NF-κB-mediated intestinal inflammation. *Journal of Ethnopharmacology*, *248*, 112302. https://doi.org/10.1016/j.jep.2019.112302

Zhao, Y., Hu, H., Jin, L., & Bo, W. (2011). Pyrolysis behavior of vitrinite and inertinite from Chinese Pingshuo coal by TG-MS and in a fixed bed reactor. *Fuel Processing Technology*, *92*(4), 780-786. https://doi.org/10.1016/j.fuproc.2010.09.005

Zhuo, B., Chen, C., Sun, T., Jiang, Z., Chen, Y., Liu, J., Nie, H., Yang, J., Zheng, X., & Lai, H. (2020). Patchouli alcohol protects against chronic unpredictable mild stress-induced depressant-like behavior through inhibiting excessive autophagy via activation of mTOR signaling pathway. *Biomedicine & Pharmacotherapy*, *127*, 110115. https://doi.org/10.1016/j.biopha.2020.110115