

Effect of Temperature Storage on Chemical Properties of Lokio Leaves (*Allium Chinense* G Don)

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ABSTRACT: Lokio (*Allium chinense* G Don) is widely used as seasoning in cooking because it contains amino acids that contribute to taste. Lokio is easily damaged during physical and chemical storage, affecting its quality. The study aims to determine the chemical changes of lokio leaves during storage. This study used a randomized design group with 3 temperature treatments, namely room temperature (30 °C), air conditioning (20 °C), and refrigerator (10 °C). Weight loss was tested using a digital scale, color was tested using a chromameter, water content was tested using thermogravimetry, fat using Soxhlet, protein using Kjeldal, and ash using a muffle furnace. Reducing sugars were tested using the DNS method, amino acids using HPLC, and volatile compounds using GC MS Headspace. The data was analyzed using ANOVA then continued with Duncan test if the treatment has a significant difference ($p < 0.05$). In the room temperature treatment, the best treatment was obtained for 2 days of storage, AC storage for 4 days, and refrigerator temperature for 14 days of storage. Of all the best treatments for each storage temperature, the best treatment was obtained in the storage treatment using AC temperature (20 °C) for 4 days. The treatment found that Lokio has a water content of 92.247%, fat of 2.47%, protein of 19.05%, ash of 14.20%, carbohydrates of 64.28%, and reducing sugar of 11.24 mg/ml. The major volatile compound content of the total storage is trimethyl sulfide and glutamic acid is the most amino acid in lokio leaves.

Keywords: Lokio, storage, chemical compound

INTRODUCTION

Lokio is a type of onion plant that is widely used by Batak tribes for cooking. Daily, lokio is used as a cooking spice because it has a delicious taste. Lokio's taste is inseparable from the content of amino acids that contribute to producing flavors such as aspartic acid and glutamic acid which produce flavors such as MSG, serine, alanine, and glycine produce sweetness. Arginine, phenylalanine, tryptophan, histidine, valine, leucine, isoleucine, and methionine produces a bitter taste (He *et al.*, 2018). Lokio has a composition of 2.2% protein, 0.3% fat, carbohydrates 13.1%, and ash 3.8% and has a sugar content of 11.51% (Li *et al.*, 1989; Shahrajabian *et al.*, 2020)

Storage is one of the post-harvest handling that has an important role in vegetable. Storage will affect the quality of vegetable and control the availability of goods in the market and consumer acceptance of agricultural commodities. Currently, the most common storage in Indonesia is using low temperatures with a temperature range of 5-16 °C Hasbiy (2015), conducted research on leek storage and got the result that leeks stored at a temperature of 15 °C using polypropylene plastic have a shelf life of 13 days with a moisture content of 88,33%.

Quality reduction in the form of damage and rotten leaves is an obstacle that is always faced during storage. Damage to lokio in the form of rotten leaves is generally affected by storage temperature. High storage temperatures can accelerate the rate of respiration in stored commodities

(Imamah *et al.*, 2016). In addition, high temperatures can also affect the content of amino acids, this is because protein as a source of amino acids will have denaturation that can cause the derivatization of amino acid residues (Sundari *et al.*, 2015). Research on the nutritional content of lokio bulbs and their properties related to health properties has been widely studied. However, the research on the changes that occur during the storage of lokio leaves and their effect on flavor has not been studied. Therefore, research is needed on the storage of lokio leaves and the chemical changes that occur during storage. This study aimed to discover the change in the quality of lokio leaves (*Allium Chinense* G Don) and their effect on the resulting flavor. The parameters tested include proximate, reduction sugars, amino acid, and volatile compounds.

MATERIALS AND METHODS

Material

The main material used in this study is Lokio (*Allium chinense* G. Don) obtained from Godean, Special Region of Yogyakarta, Aquadest from CV. Progo Mulyo, H₂SO₄ from Merck, catalyst, 0.1 N HCl from Mallinckrodt, Merck NaOH, 4% Merck HBr, methylene blue indicator, Merck petroleum ether, DNS from Sigma, Merck KNa Tartrate, Merck Na Metabisulfite, glucose standard from Merck, BCG MM from Merck and Phenol from Merck.

Method

Sample preparation

The lokio leaves were cleaned and separated from the bulbs. It was packaged in a plastic container covered in black plastic and stored at different temperatures, room temperature (25-30 °C), air-conditioned temperature (15-20 °C), and refrigerator temperature (5-10 °C). After being stored at each temperature, lokio leaves are dried until their water content is $\pm 2\%$ in order to be durable when its analyzed.

Proximate analysis

Analysis of proximate was done by AOAC method (AOAC, 1990).

Reduction sugar analysis

The reduction of sugar content was analyzed using the DNS method by making a DNS solution. DNS reagent was made by dissolving 3.5 dinitrosic acid by 1.06 g and NaOH by 1.98 g into 141.6 ml of an aquadest, then added 30.6 g of sodium potassium tartrate, 0.76 ml of phenol, and 0.83 g of sodium metabisulphyte, then the solution is stirred until evenly soluble. Determination of sugar content is done with 1 ml of hydrolyzed sugar solution inserted into the test tube, added with 3 ml of DNS reagent, then heated to a boiling water handler for 5 minutes, and cooled at room temperature. The solution is transferred into a cuvette, with absorbance measured at a maximum wavelength of 550 nm. Sugar levels are determined using the standard solution absorbance regression equation.

Identification of amino acids

Total amino acid testing was conducted at LPPT UGM with LC-MS by weighing samples of ± 0.1 grams and then inserted into a 50 ml thread tube test tube and then added HCl 6N as much as 10 mL and hydrolyzed in an autoclave with a temperature of 11 °C for 12 hours. The hydrolysis result was then neutralized with NaOH 6N, adding up to 150 ml. Then filtered with a filter of 0.22 μ l, Diluted with H₂O 5 times, and injected into LCMS 5 μ l. The mobile

phase that was used is A: 0.1% Pentadecafluorooctanoic Acid (PDFOA) 99.5%:0.5% Water/CH₃CN with 0.1% Formic acid, B: 0.1% PDFOA, 10 %:90% Water/CH₃CN with 0.1% Formic acid with an alir rate of 0.6mL/min.

Identification of volatile compounds

Analysis of volatile compounds content was carried out at the LPPT Universitas Gadjah Mada using the GC-MS Headspace method, each sample was entered in a Sample bottle and ready to be injected in GCMS with Column specifications: HP-5MS UI, Length 30 m, ID 0.25 mm, Film 0.25 μ m, Max Temp: 325/350 °C. GC MS conditioned with UHP Helium Gas Carrier (He), Injector Temperature 200 °C, Agitator Temperature 150 °C, Incubation 15 minutes, Syringe Temperature 150 °C, Split flow 20 ml/min Split ratio 20, Front Inlet Flow 1.00 mL/min, MS transfer line temp 230 °C. Ion Source temp 230 °C, Mass Range 25-350 (AMU), Purge Flow 3 mL/min, Gas Saver Flow 5 ml/min, Gas Saver Time 5 minutes.

RESULT AND DISCUSSION

Proximate contents in Lokio leaves during storage

The results showed that storage treatment significantly affected water, fat, and protein content and had no significant effect on ash and carbohydrate content (Table 1). According to Singh and Sagar (2010), packaged vegetables have an increase in water content during storage and an increase in room temperature was relatively higher than low temperature. This occurs because the amount of water metabolites as a by-product of the respiration process is higher than the water that evaporates through the transpiration process, resulting in water accumulation between cells. Therefore, Lokio leaves are susceptible to rot because of their high-water content. Low water content in effective ingredients has a long shelf life because it does not allow microbes to grow and develop so that damage can be delayed (Widaningrum *et al.*, 2010)

Table 1. Chemical Properties of Lokio Leaves During Storage at Different Temperatures

Treatment temperature	Day	Water content (%wb)	Fat (%db)	Protein (%db)	Ash (%db)	Carbohydrate (By different) (%db)
Room (25-30 °C)	0	88.11 \pm 0.033 ^a	2.95 \pm 0.169 ^c	20.90 \pm 0.001 ^c	14.85 \pm 0.066 ^a	61.28 \pm 0.068 ^a
	1	89.62 \pm 0.002 ^{ab}	2.65 \pm 0.077 ^b	19.37 \pm 0.004 ^b	14.70 \pm 0.028 ^a	63.23 \pm 0.024 ^a
	2	90.82 \pm 0.001 ^{ab}	2.45 \pm 0.227 ^{ab}	18.73 \pm 0.004 ^b	14.70 \pm 0.003 ^a	64.09 \pm 0.003 ^a
	3	92.66 \pm 0.022 ^b	2.25 \pm 0.102 ^a	17.77 \pm 0.007 ^a	14.45 \pm 0.015 ^a	65.51 \pm 0.017 ^a
AC (15-20 °C)	0	90.16 \pm 0.012 ^a	2.82 \pm 0.073 ^a	20.25 \pm 0.002 ^b	14.23 \pm 0.003 ^a	62.75 \pm 0.032 ^a
	2	91.25 \pm 0.001 ^a	2.66 \pm 0.066 ^a	20.03 \pm 0.007 ^b	14.21 \pm 0.010 ^a	63.16 \pm 0.004 ^a
	4	92.25 \pm 0.009 ^a	2.47 \pm 0.022 ^a	19.05 \pm 0.006 ^{ab}	14.20 \pm 0.002 ^a	64.34 \pm 0.005 ^a
Refrigerator (5-10 °C)	6	93.27 \pm 0.046 ^a	2.19 \pm 0.183 ^a	18.06 \pm 0.014 ^a	14.85 \pm 0.066 ^a	65.55 \pm 0.005 ^a
	0	89.29 \pm 0.023 ^a	2.89 \pm 0.136 ^a	21.74 \pm 0.002 ^b	15.24 \pm 0.038 ^a	60.10 \pm 0.035 ^a
	7	91.50 \pm 0.014 ^a	2.56 \pm 0.038 ^a	20.65 \pm 0.007 ^{ab}	15.04 \pm 0.016 ^a	61.71 \pm 0.020 ^a
	14	92.26 \pm 0.015 ^a	2.36 \pm 0.121 ^a	19.60 \pm 0.004 ^a	14.96 \pm 0.005 ^a	63.05 \pm 0.011 ^a
	21	93.36 \pm 0.046 ^a	2.12 \pm 0.015 ^a	18.73 \pm 0.018 ^a	14.92 \pm 0.018 ^a	64.20 \pm 0.014 ^a

This decrease is thought to be because, during storage, fat is oxidized to free fatty acids. With the oxidation of fat, its content will decrease. The decrease in fat content during storage is possible because fat decomposition occurs due to the oxidation process by air or oxygen which occurs spontaneously because the material is left in contact with air (Ketaren, 2005).

The decrease in N levels can also be caused by the decomposition process in lokio leaves. This indicates that the presence of microorganisms plays a role in decomposing lokio leaves. The decrease in total N levels is also thought to be due to evaporation during transpiration (Sunjoto *et al.*, 2014).

The ash content of lokio leaves does not significantly decrease because influenced by the use of minerals to sustain the life of microorganisms, because microorganisms need minerals to maintain their lives even in small amounts. Differences in the level of ash contained in materials are caused by minerals in the material, soil, and air contamination during processing (Ridal, 2003). Most of the food is 96% made up of organic matter and water, while the rest consists of mineral elements. Minerals in food are usually determined by intoxication. Sudarmaji (2007) states that ash content is a component that contains mineral elements that are left behind after the material is burned to be free of carbon. This component is not volatile in the process of burning and spawning organic compounds.

From some of these storages, it is known that storage at low temperatures is able to maintain carbohydrate content longer than high-temperature storage, this is because at low-temperature storage the respiration rate is slower so the breakdown of glucose as a carbohydrate component is also slow.

Reducing sugar

The process of heating or storage for a long time will form a brown color. The content of reducing sugars in foodstuffs produces a brown color that is expected and important for food. Generally, browning in heated or stored food will experience a reaction between reducing sugars (e.g. D-glucose) with free amino acids or free amino groups of amino acids which are part of the protein chain. This non-enzymatic browning reaction is called the Maillard reaction, this reaction plays an important role in the formation of color, aroma, and flavor (BeMiller, 1996).

Reducing sugar is important and plays a critical role in developing flavor and color in food products through non-enzymatic browning reactions. Reducing sugar reacts with amino acid when heated above 110 °C, giving a volatile aroma compound. Lokio leaves most applied at high temperatures to season a food. The results showed that the reducing sugar content increased during storage

and then decreased after some time of storage, this indicates that the storage time is directly proportional to the reducing sugar content (Figure 1). The longer the storage time of dahlia tubers, the more sugar reduction content will increase. This is because complex carbohydrates in tubers are converted into simpler components during the storage process. According to Sukmawati (1987), during storage, the carbohydrates in the tubers will be broken down into simple sugars used as a substrate during respiration. The longer the storage process, the reduced sugar content of dahlia tubers will increase. This is because polysaccharide compounds such as inulin are degraded during respiration and produce simple sugars. This is supported by the opinion of Sentana (1994) which states that the increase in reducing sugars is due to the breakdown of starch which becomes simple sugars that will be used as respiration substrates.

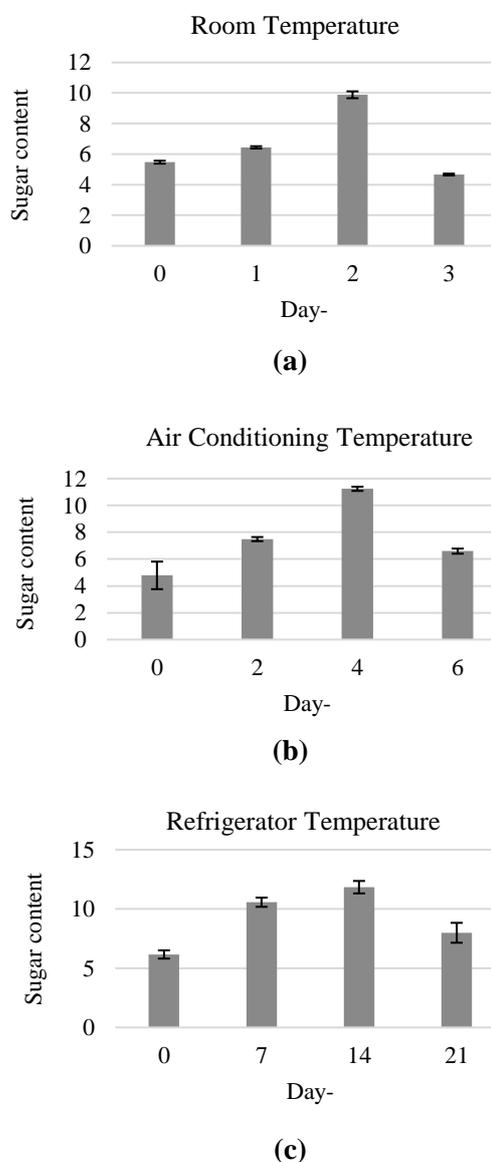


Figure 1. (a) reducing sugar content at room temperature; (b) reducing sugar content at air conditioning temperature; (c) reducing sugar content at refrigerator temperature

After the tubers are harvested, respiration continues so the conversion of starch into simple sugars will continue. During storage, tubers with high starch content will have a decrease in starch content and an increase in sugar content, because the starch contained in the form of food reserves in cells or tissues is converted into simple sugars. But the sugar content will decrease due to the decay of the material which is caused by the increasing number of microbes that break down glucose. This is in line with the research of Simbolon (2008) on the addition of tape yeast which causes the number of microbial reformers contained in the tape, especially the invertase enzyme produced is increasing, so that more glucose is converted into ethanol and consequently, the reducing sugar content decreases.

Amino acids

Amino acid profiles can provide important information regarding the composition of essential and non-essential amino acids in addition to indicating the overall composition of amino acids which can affect the taste characteristics of the sample being analyzed (Pratama *et al.*, 2017).

From Table 2. it was found that the most free amino acid content contained in Lokio during storage at AC temperature (15 – 20 °C) was Glutamic Acid. The highest level of free glutamate in Lokio is 9.53% w/w. Based on its TAV value, glutamate has the highest value compared to other amino acids, which is 9.53% w/w. It means that glutamate is the strongest influence on the taste of Lokio.

The taste sensation of glutamate is umami. The umami taste is also produced from the aspartate.

High levels of glutamate cause lokio leaves to have an umami taste which can potentially be developed into a flavoring. In addition, several amino acids have decreased, such as aspartic acid, glycine, threonine, arginine, tyrosine, methionine, valine, and leucine. This can be caused by the use of storage temperatures where according to Safaryani *et al.* (2007). Amino acids are rapidly reduced during low-temperature storage between 6-20 °C but stable at 2 °C. The presence of high glutamate in free amino acids does not necessarily serve as a parameter for Lokio as a source of umami. This is because amino acids are not the only component of taste. The taste components include D'Nucleotides, dissolved sugars, and minerals.

Volatile compounds

From the results of research on Lokio leaf storage, it was found that the most abundant component of Lokio leaves is Methanethiol (Thiol) which gives a sensation of smell like rotten cabbage (O'neil, 2006) and an unpleasant taste of cabbage (Furia, 1980), Butanal, 3-Methyl (aldehyde) which gives a warm, herbaceous, slightly fruity, and nut-like taste sensation (Furia & N. Bellanca, 1975) and an apple-like odor (Lewis, 2007), Butanal, 2-Methyl (aldehyde) with an apple-like odor sensation (Lewis, 2007), powerful penetrating, pungent odor at low levels, the resulting flavor is warm, herbaceous, slightly fruity, and nutty (Furia & Bellanca, 1975).

Table 2. Free Amino Acids contents of lokio at AC storage (15-20 °C)

Amino acid	Content (% w/w) AC storage		Taste ^{*)}	Threshold (mg/ml)	TAV	
	Day 0	Day 2			A0	A2
Aspartic acid	1.48	1.45	Umami	1.00	1.48	1.45
Glutamic acid	2.62	2.86	Umami	0.30	8.73	9.53
Serine	0.76	0.79	Sweet	1.50	0.50	0.52
Glycine	0.84	0.81	Sweet	1.30	0.64	0.62
Threonine	0.90	0.86	Sweet	-	-	-
Alanine	1.15	1.15	Sweet	0.60	1.91	1.91
Lysine	0.71	0.72	Sweet	0.50	1.42	1.44
Histidine	0.26	0.28	Bitter	0.20	1.3	1.4
Arginine	0.81	0.76	Bitter	0.50	1.62	1.52
Tyrosine	0.91	0.71	Bitter	-	-	-
Methionine	0.19	0.14	Bitter	0.30	0.63	0.46
Valine	0.92	0.90	Bitter	0.40	2.3	2.25
Phenylalanine	0.86	0.87	Bitter	0.90	0.95	0.96
I-leucine	0.63	0.65	Bitter	0.90	0.7	0.72
Leucine	1.32	1.30	Bitter	1.90	0.69	0.68
Total	14.37	14.24				

*) based on Mouritsen dan Styrbæk (2014) research.

Disulfide, Dimethyl (sulfide) with an onion-like odor sensation, sulfurous (Macbean, 2008) a pungent onion smell (Burdock, 2004), Dimethyl trisulfide (sulfide) with a sulfur-like odor sensation, alliaceous, cooked, savory, meaty, eggy and vegetative with a fresh, green, onion and sulfur taste sensation, alliaceous, gas, savory with fresh vegetative nuances (TheGoodscentcompany, 2021).

According to Wang (2019), decreasing methyl cysteine sulfoxide content of onions during storage is an important non-amino acid of vegetables belonging to the Allium genus. Meanwhile, Methanethiol (Thiol), 2 methyl propanal, and 3 methyl butanal increased. According to Shinha (2018), overall VOC emission levels from

propene, carbon disulfide, isoprene, pentane, 2-methyl furan, 3-methyl furan, 1-propanetriol, hexane, and methyl propyl disulfide increased during storage due to aging and spoilage due to the presence of microbes.

The number of dominant volatile compounds detected at room temperature (25-30 °C) (Table 3). This is thought to be caused by tissue damage that occurs at room temperature (25-30 °C) which is higher than the damage that occurs at AC temperature (15-20 °C) (Table 4) and refrigerator temperature (5-10 °C) (Table 5). In addition, according to Wang *et al* (2019), more VOCs are produced at high temperatures compared to low temperatures.

Table 3. The Dominants Volatile compounds of lokio at room temperature (25-30°C)

No	Volatile compound	Day 0		Day 1		Day 2		Day 3	
		RT (min)	Area (%)						
1	2-Aziridinylethyl Amine	1.44	6.25	1.44	5.93	-	-	1.44	5.06
2	Acetaldehyde	1.49	7.73	-	-	-	-	1.49	6.55
3	Methanethiol	1.53	13.75	1.52	13.13	1.53	11.01	1.51	10.81
4	Propanal	1.65	6.79	1.65	7.02	1.65	6.24	1.65	5.79
5	Propanal, 2-Methyl-	1.86	5.89	1.86	7.70	1.86	8.13	1.86	10.57
6	Butanal, 3-Methyl-	2.49	5.08	2.48	6.57	2.48	6.34	2.48	9.41
7	Butanal, 2-Methyl-	2.58	5.71	2.58	7.85	2.57	7.37	2.58	11.09
8	Disulfide, Dimethyl	3.64	17.99	3.64	17.56	3.63	17.88	3.64	15.71
9	Tricyclo[4.3.1.1(3,8)] Undecan-1-Amine	7.52	5.39	-	-	-	-	-	-

Table 4. The Dominants Volatile compounds of lokio at AC temperature storage (15-20°C)

No	Volatile compound	Day 0		Day 2		Day 4		Day 6	
		RT (min)	Area (%)						
1	Pyrimidine- 2,4(1H,3H)-dione, 5-amino-6-nitroso-	1,49	9.88	1.49	8.61	-	-	1.49	7.51
2	Methanethiol	1.53	6.86	1.52	12.74	1.55	6.64	1.56	7.70
3	Propanal, 2-methyl-	1.86	7.40	1.86	7.33	1.86	9.41	1.86	9.90
4	Butanal, 3-methyl-	2.48	7.51	2.48	7.28	2.49	7.82	2.49	7.60
5	Butanal, 2-methyl-	2.57	7.05	2.58	7.44	2.58	9.70	2.58	10.06
6	Disulfide, dimethyl	3.63	13.79	3.64	17.08	3.65	16.16	3.66	16.66
7	(E)-1-Methyl-2-(prop- 1-en-1-yl) disulfane	8.18	5.38	8.20	0.89	8.20	0.58	8.20	0.41
8	Dimethyl trisulfide	9.05	17.42	9.05	7.91	9.05	8.58	9.05	8.60

Table 5. The Dominants Volatile compounds of lokio at Refrigerator temperature storage (5-10 °C)

No	Volatile compound	Day 0		Day 7		Day 14		Day 21	
		RT (min)	Area (%)						
1	Glycidol	1.49	5.64	-	-	-	-	-	-
2	Methanethiol	1.52	7.74	1.51	3.53	1.51	11.32	1.52	10.48
3	Butanal, 3-methyl-	2.49	12.55	2.49	11.06	2.49	11.99	2.49	10.30
4	Butanal, 2-methyl-	2.58	13.38	2.58	12.29	2.58	13.74	2.58	14.05
5	Disulfide, dimethyl	3.65	12.25	3.64	11.50	3.64	14.11	3.64	13.47
6	Dimethyl trisulfide	9.04	7.08	9.05	10.59	9.04	5.89	9.02	1.88

This is due to the higher than at lower temperatures. This enzyme degrades S-alk(en)yl cysteine sulfoxide into acetone, acetic acid, dimethyl disulfide, and other sulfur compounds such as sulfenic acid, monosulfide, disulfide, trisulfide, and tetrasulfide in onion tubers and other vegetables of the genus *Allium*. Overall, the most identified volatile compounds were found at refrigerator temperature (5-10 °C), AC temperature (15-20 °C), and room temperature (25-30 °C). This is thought to occur due to differences in the vapor point of each compound which allows volatile compounds to evaporate at temperatures below 30 °C.

The longer the sample is stored, the less volatile compounds can be detected, this can be due to changes in the chemical content of the main ingredients, the water content, which according to Downes *et al* (2009) in the research using shallot samples, the longer onion bulbs stored have higher levels of increasing water. This increase in water content causes the components of the onion aroma to decrease. The availability of water in the tubers is a factor in the concentration of the flavor and aroma components of onions. Volatile levels in shallots change during storage. This is because the amount of alliin compound that causes the distinctive smell of shallot during storage has changed. The distinctive aroma of shallots is influenced by the activity of the alliinase enzyme, which increases with the activity of the enzyme in the tissue and vice versa, decreases with the decrease in the activity of the enzyme (Downes *et al.*, 2009). Therefore, more volatile compounds were detected in fresh ingredients than after storage.

The formation of flavors from aldehydes, ketones, alcohols, esters, and acids from fatty-oil components in plant organic matter, naturally may occur, especially during storage, namely through hydroxyacid cleavage reactions to form lactone compounds or beta-oxidation reactions and/or oxidation reactions catalyzed by lipoxygenases (Reineccius, 1994). Lokio have been widely used for their culinary and medical aspects and may reflect their effects on the olfactory, gustatory, and somatosensory systems. Plants from *Allium spp* are known to have a volatile component in the form of sulfur which contributes to the flavor and is responsible for the aroma or odor produced. In particular, biologically active components such as organo sulfuric, S-alk(en)yl-L-cysteine sulfoxides such as alliin and -glutamylcysteine dominate the flavor components of shallots and garlic. In intact cells, sulfoxides are abundant in the cytoplasm and vacuoles. Damage to plant parts results in hydrolysis of alk(en)yl-cysteine sulfoxide which then forms volatile alk(en)yl-thiosulfinates such as alliin and lipid-soluble sulfur compounds such as diallyl sulfide, diallyl disulfide which are the principal flavor compounds. Alliin is the main component of thiosulfinate which is formed due to alliinase activity in alliin (Gitin *et al.*, 2014). According to Bautista *et al.*, (2005) alliin and diallyl disulfide are

the two main ingredients in onions that can directly activate Transient Receptor Potential Ankyrin 1 (TRPA1) in trigeminal ganglia sensory neurons.

CONCLUSION

Room temperature (25-30 °C) storage resulted in the best treatment on the second day of storage, while the AC temperature (15-20 °C) on the 4th day and the refrigerator temperature (5-10 °C) on the 14th-day storage. From the chemical components, the best result was found at AC temperature (20 °C) for 4 days. In the treatment, it was found that Lokio has a water content of 92.247%, fat 2.468%, protein 19.045%, ash 14.20%, carbohydrates 64.28%, reducing sugar 11.24% maintain amino acid content, and has a more dominant volatile compound than others. Hence, it is recommended to store lokio leaves at a temperature below 20 °C not more than 4 days to maintain their quality.

SUGGESTION

This research needs to develop in testing D'Nucleotides, dissolved sugars, and minerals to give better flavor test.

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