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Antioxidant, Discoloration, Reducing Sugar, Free Amino Acids Profiling Of Black Garlic Processed On Electric Rice-Cooker

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***Abstract*— Black garlic is garlic processed by heating at a controlled temperature and humidity. The heating process of black garlic is garlic processed by heating at controlled temperature and humidity. Black garlic can be prepared in an electric rice cooker, but information is limited. The purpose of this study was to determine the color, pH, water content, antioxidant activity, flavonoids, phenolics, reducing sugar, and free amino acids of black garlic that were processed in an electric rice cooker during the heating time. The production of black garlic in this study uses an electric rice cooker. As for heating black garlic with an electric rice cooker using a temperature of 71-74 oC, RH 44-48%, and for 24 days. Heating time used for black garlic production was 0, 6, 12, 18, and 24 days. This study used a randomized block design. Black garlic cooked in an electric rice cooker during the heating time showed a darker color change towards browning and decreased water content and pH. Antioxidant activity, total flavonoids, phenolics, reducing sugar and free amino acids black garlic increased and 24 days heating time had the highest antioxidant potential. The Maillard reaction of black garlic has a higher antioxidant potential and darker discoloration than garlic.**

***Keywords*— antioxidant; black garlic; electric rice cooker; free amino acids ; reducing sugar**

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I. INTRODUCTION

Black garlic is garlic that is processed by heating by adjusting the temperature and humidity of the room, which is controlled, and the processing time is approximately one month [1]. Black garlic has advantages that can be applied in the health and food fields, especially as an antioxidant and flavor enhancer. Consuming black garlic regularly can prevent complications of type 2 diabetes [2]. Black garlic has stronger antioxidant activity than garlic. The amount of S-allyl-cysteine in black garlic is five to six times higher than garlic [3,4].

One of the primary sulfur-containing amino acid molecules believed to be in charge of garlic's advantageous properties, such as its antioxidant, anticancer, and neurotropic properties, is s-allylcysteine (SAC) [3,5]. Flavonoids, a group of phenolic compounds found in many plant tissues, can act as antioxidants [6].

In black garlic, a non-enzymatic browning reaction known as the Maillard reaction occurs. The Maillard reaction contributes significantly to aroma, taste, and color, as well as antioxidant potential [7]. During the heating process, the color changes from white to blackish brown. The process of making black garlic generally uses a thermohygrostatic

chamber, oven, and so on. Garlic is heated to 70 degrees Celsius in an oven and left to stand for up to 35 days to make black garlic. But you can also make black garlic on your own using an electric rice cooker [8].
Electric rice cookers are cooking tools that are in great demand by many people because they offer convenience and practicality in cooking, cooking, and heating rice. The electric rice cooker is on the 'warm' setting, around a temperature of 70 oC to 76 oC. An electric rice cooker has the potential as a tool that can be used to produce black garlics, such as thermohygrostatic with a temperature of 70 oC. However, information about the characteristics of black garlic processed with an electric rice cooker is still limited. Making black garlic based on [1] was by heating garlic in a thermohygrostatic chamber at 70°C and 90% RH observed for 35 days. In this study, the highest antioxidant activity occurred on the 21st day of heating, but there was a significant decrease until the 35th day. The length of heating time also gives different characteristics of black garlics.
Garlic contains free amino acids and reducing sugars. Free amino acids found in garlic are alanine, glycine, valine, leucine, isoleucine, threonine, serine, proline, seistein, asparagine, asparic acid, glutamic acid, phenyl alanine, glutamine, lysine, tyrosine, and tryptophan [9]. Zhang et al. [10] stated that the reducing sugar content in garlic was 5.80 g/kg and black garlic was 128.50 g/kg with heating for six days. The browning of garlic into black garlic is a non-enzymatic browning reaction known as the Maillard reaction. Variables in the Maillard reaction include substrate (reducing sugars and free amino acids), pH, aw, and temperature [11]. The Maillard reaction is a non-enzymatic browning interaction between reducing sugars and amino acids, peptides, or proteins that produces various products, making a significant contribution to the aroma, taste, and color, as well as to the antioxidant potential of stored and processed foods [7].
The purpose of this study was to determine the color, pH, water content, antioxidant activity, total flavonoids, total phenolics, reducing sugar, and free amino acids of black garlic processed in an electric rice cooker during the heating time. The use of different heating times in black garlic production is 0, 6, 12, 18, and 24 days. Information about black garlic, which is processed with an electric rice cooker, is expected to increase public interest in consuming black garlic given its antioxidant potential. In addition, it also provides information on how high its antioxidant potential is when compared to black garlic processed by other methods.

II. MATERIAL AND METHODS

A. Material

Sample material used in this study was garlic (Allivum sativum) obtained from Klaten Market, Central Java, Indonesia. Black garlic is processed in an electric rice cooker. Black garlic is processed in an electric rice cooker under warm conditions (± 71-74 oC, RH 44-448%). Heating for black garlic production was carried out at different times for 0, 6, 12, 18, and 24 days. The chemicals and reagents used were Aquadest, Methanol (Merck), Aluminum Chloride Hexahydrate (Merck), Quercetin Standard (Sigma Aldrich), Folin Ciocalteu (Merck), Gallic Acid (Sigma Aldrich), Na2CO3 7% (Merck), DPPH (Sigma Aldrich), Dinitrosalicylic Acid (Sigma Aldrich), Potassium Ferrocyanide (Smart Lab), Zinc Acetate Solution (Merck), Standard Glucose (Sigma Aldrich), Phosphate Buffer (Merck), Ninhydrin Solution (Smart Lab), Lysine (Merck).

B. Methods

Determination of color
Color determination was carried out with a Minolta chromameter CR-400, Konica Minolta Optics. The color measurement results obtained L*,a* and b* data. The L* value of the brightness indicator goes from black (L*=0) to white (L*=100). The a* value of the greenness to redness indicator is from -60 (green) to 60 (red) and b* the blueness to yellowness indicator is −60 (blue) to 60 (yellow). The total color difference (ΔE) is calculated by:

$$\Delta E = \sqrt{((\Delta+L^*)2 + (\Delta a^*)2+(\Delta b^*)2)}$$

ΔL*, Δa* and Δb* are the differences between the black garlic and garlic samples (day 0) at the respective L*, a* and b* values [10].

Determination of water content with a moisture analyzer
The black garlics were sliced, and then the water content of the black garlics was determined by measuring the weight using a moisture analyzer from Ohaus instruments. As much as 0.6 g of black garlic and the gar bulbs were dried at 105 oC [1].

Determination of pH using a pH meter
As much as 10 g of black garlic was dissolved in 100 mL of distilled water and filtered through 1 mm filter paper [1]. The pH of the black garlic sample was then measured using a pH meter using pH Lab 860 BNC Set with ILA170.

Antioxidant sample extraction
Sample Extraction 1.5 g of garlic was extracted with 10 mL methanol and stirred for one hour. Then do ultrasonic vibration for 20 minutes. Then the extract was centrifuged with a Zenithlab centrifuge (LC-05B) at 3000 rpm, and the supernatant was filtered. The extraction procedure was repeated one more time, and the supernatant was collected. To standardize the concentration of 1 mg/mL, the dry matter weight was determined gravimetrically [12].

Determination of total flavonoids
Determination of total flavonoids by a method based on aluminum chloride calorimetry. A total of 0.5 mL of extract sample was taken, and 0.5 mL of AlCl3.6H2O solution was added. Then vortexed and incubated at room temperature for

60 minutes. Absorbance measurement of quercetin standard solution with a 420 nm UV-VIS spectrophotometer B-ONE UV-VIS 100 DA-X spectrophotometer [13].

Determination of total phenolic
Merck EMD millipore by the Folin-Ciocalteu method using gallic acid as standard. A total of 125 µl Folin Ciocalteu (Merck Sigma Aldrich), 125 µl sample, and 250 µl distilled water were put into a test tube. Then the mixture was vortexed and incubated for 5 minutes at room temperature. Furthermore, 1.25 mL of Na2CO3 7% was added, followed by the addition of 1 ml of distilled water. The mixture was vortexed and incubated at room temperature for 90 minutes. The total amount of phenolic was determined using a UV-VIS spectrophotometer at λ 756 nm [14].

Determination of antioxidant activity
Determination of antioxidant activity using the DPPH method with minor changes. A total of 1 mL of sample solution with a concentration of 1 mg/mL was added to 1.5 mL of 0.126 mM DPPH. Furthermore, the mixture was heated for 40 minutes at room temperature and in the dark. Absorbance was measured with a spectrophotometer λ 515 nm, expressed as % RSA [12].

Determination of reducing sugar
Reducing sugars are determined using the DNS (dinitrosalicylic acid) method. Sample extraction 5 g black garlic was dissolved in 100 mL of distilled water, and then the filtrate was transferred to a 100 mL measuring flask containing 5 mL of potassium ferrocyanide solution (106 g L−1, w/v) and 5 mL of zinc acetate solution (219 g L−1, w/v). Next, the mixture was extracted using ultrasonics for 30 minutes. Then the solution is left for a few minutes, filtered, and analyzed. A total of 0.8 mL of sample was added to the test tube, and 0.6 mL of dinitrosalicylic acid reagent was added. The solution was vortexed and then boiled in boiling water for 15 minutes. Next, 9.5 mL of distilled water was added after it had cooled and then vortexed. Measurement of the absorbance of standard glucose solutions with a UV/VIS spectrophotometer at a wavelength of λ 500 nm.

Determination of free amino acids
Sample extraction 1 g of black garlic was extracted with distilled water, then the filtrate was transferred to a 100-mL measuring flask and then filtered. Determination of free amino acids:ooled to room temperature and added distilled water up to 25 ml. a totalpH 8,1 mL of sample solution, 0.5 mL of phosphate buffer solution pH 8 and 0.5 mL of ninhydrin solution were placed in a 25 ml measuring flask. The solution is boiled in boiling water for 15 minutes. Then cooled to room temperature and added distilled water up to 25 ml. The absorbance of tnm, solution was measured using a UV/VIS spectrophotometer at 570 nm and lysine was used as a standard.

Statistical analysis
Statistical analysis was performed using the Statistical Analysis System (SPSS, Version 14). The data obtained was statistically analyzed using the one-way analysis of variance (ANOVA) method; if there are differences between treatments, then proceed with a significant difference test using Duncan's Multiple Range Test (DMRT) at p < 05.

III. RESULT AND DISCUSSION

A. Black garlic color value
The measurement of the color value of black garlics is expressed in the total color difference (ΔE) and the results in of measuring the color of black garlics **Fig. 1.**

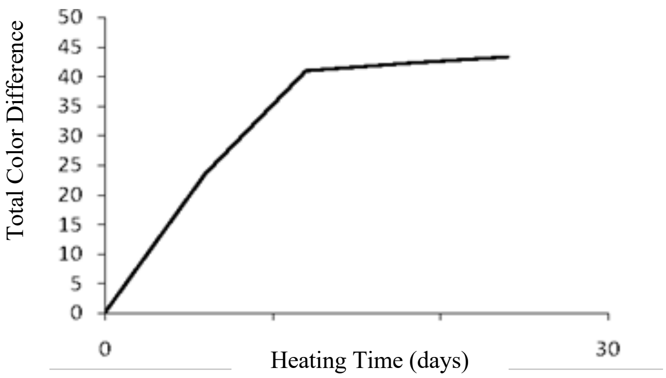

Fig. 1 Different Of Color (ΔE) Black Garlic During Heating

During the heating time, there was an increase in the total color difference (ΔE) at black garlic processed with an electric rice cooker. Increase in the total color difference (ΔE) becoming darker (darkness), then getting very dark towards browning over time. During the 6th day of heating, there was an increase in the difference in the color of the garlic, becoming darker and lighter brown in color. On the 12th day of heating, black garlic is darker than the 6th day and has a dark brown color. On the 18th day of heating, black garlics are dark brown, and on the 24th day, black garlics are slightly black in color.
Choi et al. [1] stated that in the early stages of the Maillard reaction, there was a white discharge arising from condensation of sugar-amines and Amadori rearrangement. The next reaction produces a yellow product through several stages. stages such as sugar fragmentation and amino acid degradation. In the final stage of the Maillard reaction, a darker change occurs with aldehyde-amine condensation, and a heterocyclic nitro compound is formed. The browning reaction of garlic into black garlic is a non-enzymatic Maillard browning reaction that is affected by temperature. The brown color occurs due to the formation of a high

molecular weight polymeric compound known as melanoidin formed by the reaction of Amadori and/or other dicarbonyl products with amino acids [15].

B. Water content and pH of black garlic

The water content and pH value decreased ith increasing heating time as shown in **Table 1**.

TABLE 1.
WATER CONTENT AND pH OF BLACK GARLIC DURING HEATING TIME

Heating Time (Days)	Water Content %	pH
0	60,43[e] ±1,04	6,52 ± 0,05 [a]
6	53,45[d] ±0,36	5,85 ± 0,03 [b]
12	38,20[c] ±1,63	4,96 ± 0,10 [c]
18	15,84[b] ±1,16	4,57 ± 0,06 [d]
24	1,29[a] ±0,19	4,31 ± 0,03 [e]

[a,b,c] Values with different letter notations indicate significant differences ($p<0.05$) based on the DMRT test

In black garlic, with a heating time of 24 days, the water content decreased up to forty-six times lower than fresh garlic (0 days heating time). However, black garlic that is processed with an electric rice cooker is processed in dry conditions. The operating conditions of this black garlic warm electric rice cooker were 71-74 oC, RH 44-448% and a moisture content of 1.29% at 24 days of heating. During the 24-day heating time in the electric rice cooker, the pH value of black garlic decreased. The decrease in the pH value of black garlic was up to one and a half times lower than that of garlic, from 6.52 to 4.31. The Maillard reaction is strongly influenced by pH [16]. Nursten [17] added that Amadori rearrangement has two formation pathways, namely at low pH via 3-deoxyosones and at high pH via 1-deoxy-2,3-dicarbonyls.

The water content in the black garlic Maillard reaction process is related to the browning intensity. The rate of formation of brown pigment is also affected by the water content of a system [18]. This can be seen in the water content of black garlic on days 6, 12, 18, and 24. The lower the water content, the darker it is towards browning.

Delgado et al. [19] stated that the intensity of browning also depends on the hydration of the media, and the maximum activity of the Maillard reaction is achieved at a moisture content between 10-15%. Black garlic on the 18th day of heating had a water content of 15.84% with a darker brown color than the 12th day black garlic. This shows that the heating time on the 18th day is the maximum activity of the Maillard reaction on black garlics processed with an electric rice cooker. A similar result was obtained by Bae et al. [4] that produce black garlic using a thermohydrostat chamber heating for 45 days also experienced a decrease in pH value. The decrease in the pH of black garlic correlated with its browning during the heating process. The decrease in black garlic pH is due to the formation of carboxylic acids. The formation of carboxylic acids results from the oxidation of the aldehyde groups in aldohexoses, the acidic components, and the reduction of basic amino acids in combination with sugars [4]. There are two types of reactions that can cause a decrease in pH during heating time, namely sugars and amines [20]. The decrease in pH is due to basic amines being converted to lower basic structures (primary aliphatic amines pyrrole), and secondly, some of the sugars are degraded to acids.

C. Total flavonoids and total phenolics of black garlic

Total levels of flavonoids and phenolics black garlic Processed With Electric Rice Cooker increased witth increasing heating time as shown in **Table 2** .

TABLE 2.
TOTAL FLAVONOIDS AND PHENOLICS CONTENT OF BLACK GARLIC DURING HEATING TIME

Heating Time (Days)	Total Flavonoids (mg QE/g)	Total Phenolics (mg GAE/g)
0	0,348 ± 0,13 [a]	1,697 ±0,13 [a]
6	0,465 ± 0,18 [a]	2,909 ±0,25 [a]
12	1,366 ± 0,14 [a]	6,585 ±1,06 [b]
18	4,875 ± 0,52 [b]	13,252 ±2,87 [c]
24	12,678 ± 2,12 [c]	42,02 ±1,61 [d]

[a,b,c] Values with different letter notations indicate significant differences ($p<0.05$) based on the DMRT test

The increase in total flavonoids was 36 times higher after the garlic was heated on the 24th day. On heating the black garlic on the 6th day, the total flavonoid content increased 1.3 times. This shows that the length of heating time for black garlic processed with an electric rice cooker has an effect on the increase in total flavonoids. A previous study also showed an increase in flavonoids due to heat treatment. The heat treatment disrupts the cell matrix, resulting in an increase in total flavonoids in pineapple juice [21].

The increase in the total phenolic content of black garlic processed with an electric rice cooker was in line with the increase in the total flavonoids, especially from the 18th day of heating. The total phenolic increase occurred significantly on the 18th heating day, while the total phenolic increased significantly on the 12th heating day. This shows that it is not only the flavonoid group that is responsible for the increase in the total phenolic content of black garlic.

Black garlic, which was processed in an electric rice cooker at a temperature of 71-74 oC and RH 44-48% during a 24-day heating time, had increased total flavonoid levels, according to the results of research by Kim et al. [13] on black garlic with ripening heating treatment. chamber 75 oC at 70%

RH for two days. Total flavonoids in the study consisted of flavanols, flavones, and flavonols. Black garlic flavanol and flavonol levels increased. The flavanol group consists of catechins, epicatechins, and epigalocatechins, while the flavonol group consists of myricetin, morin, and quercetin. Flavonoid compounds in black garlic such as catechins, epicatechins, and epigalocatechins from myricetin, morin, and quercetin act as antioxidants Sanbongi et al. [22] stated that catechins and epicatechins are potential candidates to act as free radical scavengers in the body and food system. The total phenolic content of black garlic is between 1.697 mg GAE/g and 42.02 mg GAE/g. The heating treatment on the 24th day produced the highest total phenolic content, which was twenty-four times that of garlic. The length of heating time affects the total phenolic content of black garlic. Bunea et al. [23] stated that the increase in phenolic acid concentration was accompanied by heat treatment because phenolic acid was released from the matrix as a result of the breakdown of the supramolecular structure containing phenolic groups. Ozcan et al. [24] stated a class of phenolic compounds, namely flavonoids, phenolic acids, tannins, and stenleid. The phenolic compounds contained in black garlic are not only flavonoids; there may also be phenolic acid, tannin, and stenleid groups.

The antioxidant potential of black garlic in the electric rice cooker method can also be seen from its constituent compounds, including phenolic compounds. The phenolic compounds seen from the total flavonoid and total phenolic content in Table 2 are associated with antioxidant activity. The higher the total flavonoid and phenolic value of black garlic, the higher the antioxidants. Flavonoids can act as antioxidants. Procházková et al. [25] stated that the antioxidant properties of flavonoids stem from their direct ability to prevent reactive oxygen species. Flavonoids also act as free radical scavengers. The process of counteracting free radicals by chelating by donating a hydrogen atom or by transferring an electron. Phenolics act as antioxidants by inhibiting enzymes involved in radical formation. Castellano [26]. Black garlic processed with an electric rice cooker has almost the same antioxidant activity, total flavonoids, and phenolics as black garlic processed with other tools.

D. Black garlic antioxidant activity

The antioxidant activity of black garlic, using the DPPH method. The antioxidant activity of black garlic Processed With Electric Rice Cooker increased with increasing heating time as shown in **Table 3.**

TABLE 3.
ANTIOXIDANT ACTIVITY OF BLACK GARLIC DURING HEATING TIME

Heating Time (Days)	% RSA (mg/ml)
0	$6{,}73^{a} \pm 0{,}53$
6	$14{,}42^{b} \pm 0{,}78$
12	$30{,}71^{c} \pm 2{,}49$
18	$61{,}15^{d} \pm 0{,}80$
24	$85{,}05^{e} \pm 1{,}61$

a,b,c Values with different letter notations indicate significant differences ($p<0.05$) based on the DMRT test

Antioxidant activity, which is the scavenging of DPPH free radicals, ranges from 6.73% to 85.057% (mg/ml). Black garlic achieved the highest free radical scavenging activity (RSA) of DPPH on the 24th day of heating, followed by the 18th day, the 6th day of the 12th day, and finally, the zero-day (garlic). The Maillard reaction on black garlic, which is characterized by a darker color change towards increasing browning, is shown in Figure 1. It is known that the more browning of black garlic, the higher the antioxidant activity Dexchand [27] stated that Maillard reaction products such as premelanoidin and melanoidin have antioxidant properties that have been successfully used in food by reducing the carbonyl compounds present in melanoidin.

The results of a study by Choi et al. [1] concerning black garlic, which was processed in a thermo-hygrostatic chamber at 70 °C and 90% RH for 21 days of heating time, had eleven times the antioxidant activity of the DPPH method higher than garlic. In black garlic, which was processed with an electric rice cooker under conditions of 71-74 oC and RH 44-448% for 24 days of heating time, the antioxidant activity was twelve times higher than that of garlic. This can provide information that black garlic processed with an electric rice cooker has antioxidant potential, especially the antioxidant activity, which is almost the same as black garlic processed with other tools. Considering the use of the electric rice cooker is very well known by the public and its practicality and convenience.

E. Reducing sugar of black garlic

Reducing sugar black garlic processed with electric rice cooker increased with increasing heating time as shown in **Table 4** .

TABLE 4.
REDUCING SUGAR OF BLACK GARLIC DURING HEATING TIME

Heating Time (Days)	Reducing Sugar (mg/g)
0	0,372[a] ±0,05
6	2,136[a] ±0,08
12	12,429[b] ±1,98
18	25,738[c] ±2,00
24	18,002[d] ±3,28

[a,b,c] Values with different letter notations indicate significant differences (p<0.05) based on the DMRT test

The reduced sugar content of black garlic processed using an electric rice cooker ranges from 0.372 mg/g to 25,738 mg/g. Table 4 shows that the reduced sugar content of black garlic from the beginning of the heating time increased until the 18th day and ended with a decrease on the 24th day. Black garlic, with a heating period of 18 days, has a reducing sugar content sixty-nine times higher than garlic. The highest reducing sugar content of black garlic was obtained on the 18th day of heating at 25,738 mg/g, followed by the 24th day at 18,002 mg/g, followed by the 12th day at 12,429 mg/g, and the 6th day at 2,136 mg/g. which is not significantly different from day 0 (garlic) of 0.372 mg/g. The reduced sugar content increases after processing garlic with an electric rice cooker. The main substrates involved in the Maillard reaction are carbonyl groups, especially from reducing sugars, and amino groups, especially free amino acids [19]. Nursten [17] stated that in the initial stage of the Maillard reaction, reducing sugars are condensed with compounds having free amino groups to give N-substituted glycosylamine condensation products, which rearrange to form Amadori rearrangement. Reducing sugars is one of the main substrates in the Maillard reaction of black garlic. However, as the Maillard reaction progressed by heating the electric rice cooker, the sugar reduction in black garlic increased until the 18th day of heating. Claude et al. [28] stated that thermal processes degrade polysaccharides into monosaccharides or oligosaccharides. During ten days of heating, black garlic polysaccharide levels decreased from 98.4% to 29.4%, disaccharides from 8.61% to 4.86%, and monosaccharides from 1.21% to 70.21% [10]. At the end of the 24th day of heating time, the reduced sugar content of black garlic processed in an electric rice cooker decreased. Research on black garlic processed using an electric rice cooker with operating conditions of 71-74 oC and RH 44-448%, had reducing sugar levels that increased and then decreased during heating in accordance with the research results of [29]. Zhang et al. [29] stated that during the processing of black garlic, the reduced sugar content in black garlic depends on two factors. Firstly, the polysaccharides in garlic are degraded into sugars, and secondly, the reducing sugars are consumed during the Maillard reaction. The Maillard reaction of black garlic, apart from producing a sour taste, also has a sweet taste. The sweet taste of black garlic increases with the longer heating time. Choi et al. [1] stated that the sweet taste of black garlic could be attributed to the increase in sugar content (glucose, fructose, glucose, maltose, and sucrose) from white garlic to black garlic. However, at the end of the heating time period, the 24th day, the sweet taste of black garlic began to decrease, and it became more sour taste, and bitter compounds began to be felt. This is supported by data on the reduced sugar content of black garlic on the 24th day of heating, which was lower, and the pH was significantly lower compared to the 18th day. Martins et al. [30] stated the Maillard reaction in the glucose/glycine system model, glucose isomerization via 1,2-enolization and 2,3-enolization. Isomerization through 1,2-enolization degrades sugars into organic acids called formic acid and C5 fragments. Formic acid is thought to be formed through 1,2-enediol from the cleavage of the C1-C2 and C5 fragments, possibly from 2-deoxyribose and 3-deoxy pentose.

F. Free amino acid of black garlic

Reducing sugar in black garlic processed with an electric rice cooker increased with increasing heating time and decreased finally at heating time, as shown in **Table 5**

TABLE 5.
FREE AMINO ACIDS OF BLACK GARLIC DURING HEATING TIME

Heating Time (Days)	Free Amino Acids (mg/g)
0	0,350[b] ±0,02
6	0,375[b] ±0,31
12	0,425[c] ±0,02
18	0,513[d] ±0,06
24	0,135[a] ±0,01

[a,b,c] Values with different letter notations indicate significant differences (p<0.05) based on the DMRT test

Free amino acid levels increased with increasing heating time and decreased at the end of heating time. This shows that the length of heating time affects the levels of free amino acids in black garlic. Black garlic, with a heating time of 18 days, has the highest free amino acid content, almost one and a half times that of garlic. The free amino acid content of garlic was 0.3504 mg/g, and for heating black garlic on days 6, 12, 18, and 24, respectively, it was 0.3751 mg/g, 0.4251 mg/g, 0.5125 mg/g, and 0.1531 mg/g. Free amino acids are one of the main substrates in the Maillard reaction. The free amino acids found in garlic are alanine, glycine, valine, leucine, isoleucine, threonine, serine, proline, asparagine, aspartic acid, glutamic acid, phenylalanine, glutamine, lysine, tyrosine, and tryptophan [9]. In the Maillard reaction of black garlic, free amino acids are one of the main substrates.

However, as the Maillard reaction progressed with heating, the free amino acids of black garlic increased until the 18th day of heating. The black garlic heating process from research by [31] also measured amino acid levels. The results of the research show that there are amino acids in black garlic, namely alanine, valine, leucine, isoleucine, lysine, arginine, proline, asparagine, aspartic acid, tyrosine, and phenylalanine. Black garlic amino acids in this study increased on the fifth day after the start of the thermal process. The increase in black garlic amino acids on the fifth day of the study was most likely due to the degradation of proteins or peptides, which may result from enzymatic hydrolysis or non-enzymatic hydrolysis. The research results of black garlic processed using an electric rice cooker with operating conditions of 71-74 oC and RH 44-448% for 24 days of heating had free amino acid levels, which increased and then decreased at the end of the heating time. This is in line with the research results of [32] that heat treatment at 90°C for 30 minutes on tualang honey bee, gelam honey bee, and acacia honey bee resulted in an increase and decrease in several free amino acids. The decrease in free amino acids is related to the denaturation process. The reaction of carbonyl groups from reducing sugars and amino acids during the thermal process also reduces free amino acids in honey. Kauzman et al. [33] stated that denaturation causes activity loss by unfolding or aggregating proteins at high temperatures. The levels of free amino acids in black garlic processed in an electric rice cooker are related to reducing sugar. **Table 4**. and **Table 5** show information that the longer the heating time, the higher the levels of free amino acids and reducing sugars, followed by a decrease at the end of the heating time. This is associated with the Maillard reaction process in black garlic, which is a reaction between the main substrate, namely free amino acids, and carbonyl groups in reducing sugars. The formation of reducing sugars in black garlic produced by electric rice cookers is faster than the formation of free amino acids. Sawai et al. [34] stated that when sweet potatoes are heated, the starch is gelatinized, and a β-amylase enzymatic reaction occurs, which produces reducing sugars, and 83ºC treatment produces the highest reducing sugars. This provides information that it is possible that the starch-breaking enzyme in black garlic has started to have an enzymatic reaction at the beginning of the heating time, considering that the operating conditions of the electric rice cooker are 71-74°C. The heating process allows reducing sugars to be formed more quickly than free amino acids in black garlic. The Maillard reaction in black garlic is a reaction between the carbonyl group of reducing sugar and the amine group of amino acids during heating. The Maillard reaction of black garlic processed in an electric rice cooker during heating is characterized by a darker color change (darkness), a decrease in pH, a decrease in water content, an increase and then a decrease in reducing sugars and free amino acids at the end of the heating time. Baxter [35] stated that the Maillard process's initial reaction involves forming Schiff's base with sugar and amino acids. This reaction is reversible; subsequent rearrangements are not reversible and result in the "destruction" of the amino acid. Schiff's base is labile to acid and is followed by acid hydrolysis, which adds free amino acid to Schiff's base. The next stage occurs the formation of Amadori Reangarment, which in the Maillard reaction of black garlic goes through the 1,2 enolization pathway. Tateyama [36] stated that the 1,2-eneminol pathway from Amadori Reangarment occurs by losing the hydroxyl group at C-3 with subsequent deamination at C-l, followed by dehydration, then 3-deoxyosone is formed, easily loses water molecules, and cyclizes to form furfural derivatives, which are carbonyl groups. After going through the Amadori rearrangement stage, the carbonyl group can then condense with the free amino group Hodge [37] stated that after Amadori rearrangement, the carbonyl group can then condense with the free amino group with the formation of aldehydes and aminoketones, known as Strecker degradation. Next, various reactions take place, ultimately forming brown nitrogen polymers and co-polymers, known as melanoidins. This is characterized by a change in the color of the garlic from black to darker (darkness) towards brownish.

IV. CONCLUSION

Black garlic that was processed in an electric rice cooker during the heating time showed a darker color change towards browning, decreased moisture content, and pH. Black garlic experienced the most discoloration with 24 days of heating. Antioxidant activity, total flavonoids, and black garlic phenolics increased, and the heating time of 24 days had the highest content. Maillard reaction black garlic, which is processed in an electric rice cooker, has a higher antioxidant potential than garlic. Reducing sugar and free amino acids of black garlic increased with increasing heating time and decreased at finally heating time.

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