Effect of heating using autoclave and microwave on the quality of rice bran as food material

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Abstract. Rice bran is an epidermis of rice produced from 10 - 12% of the rice milling process. Rice bran is susceptible to fat damage due to enzymatic and oxidative processes that cause rancidity, therefore a further process such as heating is needed for stabilization. This study is using the factorial Randomized Block Design (RBD) method, the variables are the type of heating method (autoclave and microwave) and heating duration. Autoclaving (30 min at 75°C) yielded best content value. The heated rice bran has 2.89% water content, 5.79% ash content, 13.87% protein, 7.24% crude fat, 2.04% free fatty acids, and 0.85% peroxides. The contents of free fatty acids, peroxide value, and proximate value from heated rice bran are better than untreated rice bran (control).

1. Introduction
Indonesia is one of the largest rice-producing countries in Asia, in addition to Bangladesh, Vietnam, Myanmar, Thailand, the Philippines, Japan, Pakistan, Cambodia, the Republic of Korea, Nepal and Sri Lanka, which produce 90% of the world's total rice [1]. Worldwide rice production, based on FAOSTAT data in 2016, reached 756 million tons [2]. In 2016, Indonesia produced 79.1 million tons rice [3]. High amount of rice production will also result in high byproducts, such as husks, coarse bran, and rice bran. Rice bran is the epidermis of the rice, located between white rice and husks, obtained as a byproduct of rice processing [4]. The rice processing into white rice requires 4 stages, including the husk separation and 3 milling stages. The first stage milling byproduct contains 10% coarse bran, meanwhile the second and third stage millings produce 10 - 12% rice bran of the total grain [5].

In general, the utilization of rice bran by Indonesian people is limited as animal feed. However, along with the technological development, people began processing rice bran into products that had more nutritional value such as pastries, cereals, probiotic drinks, and essential oil. Nutritious components in rice bran include carbohydrates, protein, fat, dietary fiber, antioxidants, vitamins, minerals, and essential unsaturated fatty acids [6], where these nutrients are good for human consumption.

Rice bran has high fat and water contents that leads to rancidity due to enzymatic and microbial activities. Those include, the formation of glycerol and fatty acids through fat hydrolysis by lipase enzyme, where the free fatty acids are further oxidized by the lipoxygenase enzyme to form peroxides, ketones, and aldehydes that give a result to rancidity [7]. Damages on rice bran can be minimized by...
several methods, such as heating with hot steam, hot air, ohmic and radiation, cooling at 4°C or modification of pH (7-8) [8]. Based on the results of free fatty acids and peroxide value analysis in several studies, heating using autoclave and microwave are effective for inactivating enzymes while maintaining bioactive components in rice bran [2,4–6,9]. Previous studies have reported that using superheated steam at 170°C for 7 min and using a microwave at 800 Watts for 3 min were effective for stabilizing the rice bran [9,10]. However, high temperature and high electrical power can cause damage to the bioactive components and nutrients in rice bran. Temperature below 100°C is sufficient to inactivate enzymes that are responsible for fat hydrolysis without damaging the nutritional value of rice bran [11]. Therefore, this study is carried out to see the effect of heating methods given by the low temperature and low electrical power with autoclave and microwave methods on bran quality, with the variation in heating duration. The bran quality is analyzed based on the content of free fatty acids and peroxides, along with proximate values of water, ash, protein, fat, and dietary fibre.

This research is expected to produce bran that meets the quality standards as food materials in functional food products. The use of rice bran in food products is important to create innovative and diverse functional food products, especially in Indonesia.

2. Methodology

2.1 Material, reagent, and instrument
The main material for this main research is bran collected from rice mills in Aceh Besar District, Aceh Province. The rice bran should be no more than 2 days after the milling process. The chemicals and reagents used for the analysis were CuSO₄, K2SO₃, Kjeldahl tablets, H2SO4, phenolphthalein, NaOH, boric acid, HCl, and filter paper. All chemicals and reagents used are pro analysis (PA) quality, purchased from Sigma Aldrich and Phum Edumedia.

2.2 Heating of rice bran
Rice bran is sifted with 180-mesh sieve before used as a sample. A total of 100 g sample is used for each heating batch. The first heating is processed with autoclave sterilizer (WAC-60 L Digital) at 75°C for 20, 30, and 40 min. The second heating uses a combination of autoclave (75°C) and microwave 180 Watt, where the sample is first inserted into autoclave for 20, 30, and 40 min, then proceed with a microwave of 180 Watt with the same duration. The last heating uses microwave (Panasonic NN-GT35HM-TTE) at 180 Watt for 20, 30, and 40 min. The combination of the experimental treatments can be seen in Table 1.

Heated and untreated rice bran are inserted into 10 × 20 cm aluminum zip lock. The package is stored at room temperature. Free fatty acids (FFA) and peroxide value (PV) content of untreated and heated rice bran are determined to saw the level of fat damage. The lowest value of FFA and PV is selected as the stable rice bran. Thereafter, stable rice bran is analyzed such as water content, ash content, protein, crude fat, and crude fiber to compare the proximate value between untreated (control) and heated rice bran.

| Types of Heating (J) | Duration of Heating (D) |
|----------------------|-------------------------|
|                      | 20 min | 30 min | 40 min |
| Autoclave (75°C)     | J₁D₁   | J₂D₂   | J₃D₃   |
| A : M (75°C : 180 W) | J₁D₁   | J₂D₂   | J₃D₃   |
| Microwave 180 W      | J₁D₁   | J₂D₂   | J₃D₃   |

Table 1. Treatment of experiment.
2.3. Analysis of rice bran
Analysis of free fatty acids (FFA), peroxide value (PV), and proximate refers to AOAC (2005). Proximate analysis was performed on the best treatment and untreated rice bran (control) of the obtained FFA and PV.

2.4. Statistical analysis
Analysis of all experiments was carried out in duplicate and the data displayed is the mean value. The data obtained are analyzed statistically using two-way analysis of variance (ANOVA) analysis. If the treatment shows an influence, then proceed with testing LSD (Least Significant Difference) at a significance level of $p < 0.05$ which can be calculated by the following equation (1).

$$
\alpha = q \sqrt{\frac{MS \text{ error}}{r}}
$$

(1)

3. Results and discussion

3.1 Free fatty acids and peroxide value of rice bran
Free fatty acid (FFA) and peroxide value (PV) are indicators to determine the level of fat damage caused by enzyme activity and lipid oxidation. The free fatty acids and peroxide value are reduced after heating treatments on the rice bran before storage, and increase during storage. The heating process reduces the fat content in the rice bran, so that the content of free fatty acids and peroxide value also decrease [6,9]. The lowest FFA and PV is considered because it indicates a low level of fat damage during storage. If the content of free fatty acids is high, then the peroxide value will also increase [2]. The results of the free fatty acids analysis in rice bran can be observed in Table 2.

Table 2. Effect of heating after treatment and after storage on FFA and PV.

| Treatments | FFA Content (%) | PV Content (meq/kg) |
|------------|----------------|---------------------|
|            | After Treatment | 45 days             | After Treatment | 45 days             |
| Untreated  | 27.63           | 58.41               | 16.13           | 28.15               |
| J1D1       | 2.88 ± 0.06$^b$ | 10.17 ± 0.2$^c$     | 1.25 ± 0.02$^b$ | 7.62 ± 0.01$^c$     |
| J1D2       | 2.04 ± 0.09$^a$ | 8.50 ± 0.04$^a$     | 0.85 ± 0.04$^a$ | 2.21 ± 0.01$^a$     |
| J1D3       | 4.58 ± 0.21$^e$ | 15.32 ± 0.4$^d$     | 2.62 ± 0.01$^d$ | 10.11 ± 0.02$^f$    |
| J2D1       | 4.11 ± 0.01$^d$ | 22.18 ± 0.1$^e$     | 2.54 ± 0.02$^d$ | 9.85 ± 0.01$^e$     |
| J2D2       | 6.47 ± 0.08$^f$ | 35.25 ± 0.1$^f$     | 3.21 ± 0.01$^e$ | 11.64 ± 0.01$^g$    |
| J2D3       | 7.35 ± 0.25$^g$ | 43.62 ± 0.02$^h$    | 4.88 ± 0.03$^f$ | 13.44 ± 0.02$^f$    |
| J3D1       | 2.16 ± 0.06$^a$ | 9.67 ± 0.01$^b$     | 1.08 ± 0.01$^b$ | 5.24 ± 0.01$^b$     |
| J3D2       | 3.58 ± 0.08$^c$ | 24.21 ± 0.1$^f$     | 1.90 ± 0.07$^c$ | 8.79 ± 0.33$^d$     |
| J3D3       | 6.16 ± 0.04$^f$ | 35.39 ± 0.03$^g$    | 3.22 ± 0.16$^e$ | 12.14 ± 0.01$^b$    |

*Values are mean ± standard deviation. Mean values followed by the same letter notation are not significantly difference ($p>0.05$).

Table 2 showed that the FFA dan PV contents in the rice bran decrease significantly after treatments. The lowest FFA and PV are shown in J1D2 (autoclaved for 30 mins) and J1D3 (microwave for 40 mins) treatments, which are 2.04% and 0.85 meq/kg respectively for J1D2, and 2.16% and 1.08 meq/kg respectively for J1D3.

These results also show that the types of heating method affect the obtained FFA and PV contents. Treatments, with the separate use of autoclave and microwave, produce rice bran with lower FFA and PV contents than the treatment with combined method. This ascribes to the higher of water content given by the combination of autoclave and microwave (J2), where water may accelerate fat hydrolysis.
and promote microbial damage such as caused by fungi and bacteria [12]. The heating using autoclaves (J1) gives the lowest increase in the formation of FFA and PV compared to others. This indicates that autoclave heating (J1) is effective to inhibit the rate of FFA and PV formation. The same result is found where low temperature and low electrical power are used. Those treatments allow bioactive compounds and nutrients to be maintained. Among the heating methods used, autoclaving is the most effective method for slowing the formation of FFA [13]. These results are comparable with the other studies reporting the use of super-heated steam at 170°C, infrared at 700 Watts, heating hot air-assisted radio frequency at 90°C, and microwave at 850 Watts, where they can reduce FFA and PV of the rice bran [2,4,9,10].

Heating duration (D) also affects the quality of the bran produced. In general, the longer the heating duration, the worse the quality of the rice bran. This can be observed by the increase of FFA and PV contents with the increasing duration. This is due to more water content trapped in the bran after a longer heating process. As previously stated, water can promote hydrolysis, oxidation, or microbial damage [2,12].

The contents of FFA and PV are found to be increasing after 45 days. This indicates that, after 45-days storage, the water content increased and triggered the formation of high FFA dan PV. This means, more FFA and PV contents are formed with a longer storage time. This is comparable with other studies where the water content in rice bran causes damage on the rice bran during storage, so that FFA and PV contents become higher [2,4–6]. In this study, the content of FFA and PV after storage are found higher compared to the other studies [2,4–6,9]. Thus, the storage optimization after the heating treatment is needed for future study.

3.2 Proximate

Proximate analysis is used to determine the chemical composition in rice bran such as water content, ash content, protein, fat, and fiber, which can indicate the quality of a food product [12]. Proximate analysis is carried out on the rice bran for the selected treatments (J1D2 and J3D1), due to the best results obtained from those treatments. Proximate analysis results show that the heating treatment affects the proximate values of rice bran samples as shown in Table 3.

| Samples | Water Content | Ash Content | Protein | Crude Fat | Crude Fiber |
|---------|---------------|-------------|---------|-----------|-------------|
| Untreated | 21.3% ± 2×10⁻⁴ | 11.7% ± 1×10⁻³ | 14.2% ± 3×10⁻⁴ | 16.1% ± 1×10⁻⁴ | 4.0% ± 2×10⁻⁴ |
| J1D2    | 2.9% ± 2×10⁻⁴ | 5.8% ± 2×10⁻⁴ | 13.9% ± 2×10⁻⁴ | 7.2% ± 3×10⁻⁴ | 2.7% ± 1×10⁻⁴ |
| J3D1    | 3.2% ± 2×10⁻⁴ | 6.1% ± 2×10⁻⁴ | 13.9% ± 1×10⁻⁴ | 7.8% ± 2×10⁻⁴ | 3.1% ± 1×10⁻⁴ |

*Values are mean ± standard deviation.

The proximate value of food determines its quality, depending on the content types of nutritional value. Indonesia has national standards (SNI 01-4439-1998) for each proximate value of rice bran. In general, the proximate value of the treated rice bran is better than the untreated rice bran. The proximate content of untreated rice bran (control) is higher. The results also show that the untreated rice bran has higher water content than J1D2 and J3D1 rice bran.

It can be seen that the heating treatment can reduce the water content of rice bran. Nevertheless, Ramezanazdeh, et al., (2000) reported that the heating process increase the bran water content in 0 days before storage. The water content in rice bran is not desirable because it cause of the low quality of bran, thus lower water content is considered better for the quality, especially for storage purposes.

Untreated rice bran also has higher fat content than the ones obtained after treatments J1D2 and J3D1. Fat is not desirable in food products because of the health concern. Excessive fat consumption
can cause increased cholesterol in the body. The heating process decreases the fat content of rice bran after heating treatment as seen in Table 3. This showed that the heating treatment is effective for reducing fat content in rice bran. However, the heating process used in this study does not significantly affect the decrease in protein content, which indicates that the heating process can well maintain the protein content.

Other studies reported that the heating process reduce the nutrient content of rice bran even though it is not significant. This decrease can occur due to the use of higher temperatures and longer duration. So that optimization is needed in future studies to determine the effects of various heating methods on the proximate value of rice bran. Therefore, rice bran with autoclave at 75°C for 30 min (J1D2) is chosen as the best rice bran because it has lower water content, ash content, and fat content than the others.

4. Conclusion
The heating method of rice bran can be done by using eco safe instruments such as autoclaves and microwaves. The quality of rice bran untreated (control) is bad, and the heating process significantly reduces free fatty acids, peroxide values, and proximate in rice bran to be better. The use of autoclave and microwave separately is better than the combination method. The best treatments for rice bran is with autoclave at 75°C for 30 min (J1D2) and a microwave at 180 Watt for 20 min (J3D1). We expect these rice bran can be the substitutes for functional food processing.

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Acknowledgements
The author would like to thank Magister program of Chemistry Department, Faculty of Mathematics and Sciences, Universitas Syiah Kuala for research laboratory and facilities.