Proximate Nutritional Evaluation of Gamma Irradiated Black Rice (*Oryza sativa* L. cv. *Cempo ireng*)

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**Abstract.** Black rice is a type of pigmented rice with black bran covering the endosperm of the rice kernel. The main objective of the present study was to provide details information on the proximate composition of third generation of gamma irradiated black rice (*Oryza sativa* L. cv. *Cempo ireng*). In respect to the control, generally speaking, there were no significant changes of moisture, lipids, proteins, carbohydrates and fibers contents have been observed for the both gamma irradiated black rice. However, the 200-BR has slightly better nutritional value than that of 300-BR and the control. The mineral contents of 200-BR increased significantly of about 35% than the non-gamma irradiated black rice.

1. **Introduction**

Rice (*Oryza sativa* L.) is an important cereal grain which consumes nearly half of the world’s population. Pigmented rice has been consumed for a long time in Asia and is considered to be a healthy food [1]. Black rice is a type of pigmented rice with dark purple of outer bran layer which appears to be black. It has high nutritional value due to its high content of fiber, minerals, anthocyanin, vitamins B and E [2-3]. This rice is low in sugar, salt and fat and free of gluten and cholesterol. In spite of its beneficial, black rice paddy has some limitations which can hinder the mass production of black rice. The demand for black rice has been increasing due to its high nutritional value and multiple biological activities. Black rice paddy has a long period of cultivation which is more than 145 days [4]. The habitus is very tall (more than 150 cm), as a result the plant falls easily.

On previous work, our group have applied gamma radiation on the black rice grains in order to overcome the limitations of the black rice [5]. The black rice grains were irradiated with gamma rays yielded by core of Cobalt 60 which gamma rays radiation doses used were 200 and 300 Gy. This irradiation was applied to induce mutation in black rice plant to improve its quality and physical appearance. This genetically engineered has been successfully modified the morphological of leaves, stems and grains of the black rice plant. After 74 days of plantation, the irradiated black rice have been flowered faster than non-irradiated one, which were 9.25 and 11.45% for 200 and 300 Gy irradiated black rice, respectively [5].
The doses of exposed gamma sources often also influence the nutrition content of cereal grains [6]. To meet the criteria on the commercial application, the irradiated products must be assayed carefully in term of safety and acceptability [7]. One of the method widely employed to evaluate the nutrition content on the product is proximate analysis following AOAC standard method [8]. The nutritional value of the irradiated black rice with doses of 200 and 300 Gy are reported in this paper.

2. Experimental
2.1. Materials
Third generation of gamma irradiated black rice (Oryza sativa L. cv. Cempo ireng) with doses of 200 and 300 Gy were used in this research. The non-irradiated black rice was applied as a control. All chemicals were used from e-Merck. Solvents of analytical grade were utilized without further purification.

2.2. Proximate Analysis
The black rice powdered was sieved through a 100 mesh sifter and stored in refrigerator for further used. Proximate analysis of black rice samples was conducted according to SNI 01-2891-1992 following AOAC 2000 [9].

2.2.1. Moisture content. The black rice samples was weighed and recorded as initial weight (Wo). Afterward, each black rice samples was dried in oven until obtaining constant weight (Wi). The moisture content was calculated according to Equation 1.

\[
\text{Moisture} = \left(1 - \frac{W_i}{W_o}\right) \times 100\% \quad \text{.................................................................} \tag{1}
\]

2.2.2. Mineral content. Mineral content of each black rice samples was determined through dry method. Black rice samples (5 g) were placed in a weighed porcelain and burned in furnace at 500-600 °C. The mineral content was expressed as the final weigh of sample.

2.2.3. Lipid content. The lipid content was examined via soxhlet extraction method. Black rice samples (5 g) were extracted using 150 mL of petroleum ether for 6 h. The solvent was then evaporated and the extract lipids were weighed (Wlipid). The lipid content was calculated using Equation 2.

\[
\text{Lipid} (\%) = \frac{W_{\text{lipid}}}{W_o} \times 100\% \quad \text{.................................................................} \tag{2}
\]

2.2.4. Protein content. The protein content was determined using Kjehdahl method. Black rice samples (0.1 g) was destructed with 1 g of CuSO₄, and 2.5 mL of H₂SO₄ for 2 h at 100 °C. Afterward, to the destructed sample was added 50 mL of distilled water and 15 mL of 50% w/w NaOH, then distilled. The distillate was collected in Erlenmeyer containing 10 mL of 0.02 N HCl. The sample was then titrated using standardized NaOH to determine protein content.

2.2.5. Carbohydrate content. The carbohydrate content was calculated using Equation 3.

\[
\text{Carbohydrate} = 100\% - \% (\text{Moisture} + \text{Mineral} + \text{Lipid} + \text{protein}) \quad \text{......} \tag{3}
\]

2.2.6. Fiber content. The fiber content was determined using hydrolysis method. Free lipid content of black rice (5 g) was hydrolyzed using 50 mL of 1.25% H₂SO₄ solution and boiled for 30 minutes. Then, 30 mL of 3.25% NaOH was added into the system and boiled for next 30 minutes. The sample was filtered in warm condition and washed with 1.25% H₂SO₄, distilled water and 96% ethanol. The sample was dried at 105 °C and weighed. The final weigh was defined as the fiber weight.
3. Result and discussion

The proximate analysis of gamma irradiated black rice is necessary to conduct regarding to the food safety and security regulation. Proximate nutritional evaluation of gamma irradiated black rice has been conducted for quantifying the moisture, minerals, lipids, proteins, carbohydrates and fibers contents. The results obtained for proximal composition of black rice investigated in this study are shown in Table 1.

Table 1. The proximate nutritional data of black rice samples.

| No. | Analysis       | Control (%) | BR-200* (%) | BR-300** (%) |
|-----|----------------|-------------|-------------|--------------|
| 1   | Moisture (%)   | 14.82       | 12.89       | 13.22        |
| 2   | Mineral (%)    | 1.23        | 1.67        | 1.13         |
| 3   | Lipid (%)      | 3.00        | 3.06        | 2.93         |
| 4   | Protein (%)    | 7.15        | 7.63        | 7.47         |
| 5   | Carbohydrate (%)| 73.80      | 74.75       | 75.25        |
| 6   | Fiber (%)      | 1.13        | 0.97        | 0.80         |

*BR-200 : gamma irradiated black rice with a dose of 200 Gy
**BR-300 : gamma irradiated black rice with a dose of 300 Gy

Generally, there is no significant nutrition content changes of gamma irradiated black rice in respects to the control one. These results were in line with other researchers. Taipina et al. (2011) reported that the macronutrient of irradiated sample is unaffected by radiation doses up to 10 kGy [7]. Moreover, Diehl and Josephson (1994) reported that the essential nutrition of irradiated samples undergo no significant losses under condition of actual or potential commercial application [10].

The moisture and fiber contents of both gamma irradiated black rice slightly decreased and moderately no changes have been observed for the lipids contents of both irradiated black rice. The moisture content of BR-200 decreased to 12.89% which is meet the acceptable of moisture content of about 12%. This moisture levels is recommended for long term storage as well as avoid microbial growth [8]. The fiber content of black rice obtained from cv. Cempo Ireng was 1.13%. Fiber hold important role on the decreasing of the blood cholesterol and sugar in diabetics, also reduce the bowel disorder as well as fight against constipation. The high content of fiber will prevent all of the mentioned diseases. However, the fiber content also should meet the required standard. Oko and Onyekwere (2010) reported that the standard fiber content of rice is 0.5% - 1.0%. Both of the irradiated black rice is still comply with the fiber content requirement which can gave a positive impact to fulfill the required fiber content to the body [11].

It is noted that the minerals content of BR-200 increased significantly of about 35% in respect to the control. However, slightly decreased in minerals content was observed for the BR-300 sample. Minerals are known as essential nutrients playing important role in the body activity [12]. Mineral content on the rice is mostly influenced by genetic characteristic and environment factors [13].

In respect to the control, the proteins and carbohydrates contents were slightly increased for both gamma irradiated black rice. Carbohydrate content which is higher than 70% can be considered to be a good source of carbohydrate [14]. Hence, these gamma irradiated black rice varieties are good source of carbohydrate. Carbohydrate in rice is primarily starch composed by amylose and amyllopectin. The carbohydrate content is strongly influence the rice physical parameter, where at high content of carbohydrate will make the individual grains is easy to sticking to each other, however at low level, it is prevented [15].

4. Conclusion

Although there were no significant changes of moisture, lipids, proteins, carbohydrates and fibers contents have been observed for the both gamma irradiated black rice in respect to the control, the 200-BR has slightly better nutritional value than that of 300-BR. The minerals, lipids, proteins and fibers contents of the 200-BR are higher than those of the 300-BR. Moreover, the minerals, lipids, proteins
and carbohydrates contents of the 200-BR is higher than those of the non-gamma irradiated black rice. It is notably increased of the minerals content of 200-BR of about 35% in respect to the control. Results generated in this study could be able to provide adequate information’s on the quality of the black rice new variant for alternative cereal grains.

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References
[1] Simachanu C S, Yenagi N B, Math K K 2016 J. Farm Sci. 29(1) 61
[2] Suzuki M, Kimur T, Yamagishi K, Shinmoto H, Yamak K 2004 Nippon Shokuhin Kagaku Kogaku Kaishi 51(58) 424
[3] Kristamtini, Taryono, Basunanda P, Murti R H, Supriyanta 2012 J. Agric. Biol. Sci. 7 982
[4] Kushwara UKS 2016 Black Rice Switzerland (SW): Springer International Publishing DOI 10.1007/978-3-319-30153-2_2
[5] Hartanti R S, Putri T A N, Zulfa F, Sutarno, Suranto 2017 IOP Conf. Ser.: Materi. Sci. Eng. 193 012038 doi:10.1088/1757-899X/193/1/012038
[6] Marcu D, Damian G, Cosma C, Cristea V 2013 J. Biol. Phys. 39 625
[7] Taipina M S, Garbelotti M L, Lamardo L C A, Santos J S, Rodas M A B 2011 Proc. Food Sci. 1992
[8] Verma D K, Srivastav P P 2017 Rice Sci. 24(1) 21
[9] AOAC International 2000 Official Methods of Analysis, 17th Ed., Washington DC
[10] Diehl J F, Josephson E S 1994 Acta Alimentary 23(2) 195
[11] Oko A O, Onyekwere S C 2010 Int. J. Biotechnol. Biochem. 6(6) 949
[12] Wang K M, Wu J G, Li G, Zhang D P, Yang Z W, Shi C H 2011 J. Cereal Sci. 54(1) 116
[13] Zimmermann M B, Hurrell R F 2002 Curr. Opin. Biotechnol. 13(2) 142
[14] Thomas R, Wan-Nadiah W A, Bhat R 2013 Inter. Food Res. J 20(3) 1345-1351
[15] Mbatchou V C, Dawda S 2013 Int. J. Res. Chem. Environ. 3(1) 308