

THE EFFECT OF BLACK RICE FLOUR AND BLACK SOYBEAN FLOUR PROPORTIONS AND SUNFLOWER SEED OIL ADDITION ON THE CHARACTERISTICS OF HIGH-ANTIOXIDANT COOKIES

*Dedin Finatsiyatull Rosida^{*1&2}, Sadrina Adsari Novita Hartono¹, Riski Ayu Anggraeni¹*

Address:

¹Department of Food Technology, Faculty of Engineering, Universitas Pembangunan Nasional Veteran Jawa Timur, Surabaya 60294, Indonesia.

²Innovation Center of Appropriate Food Technology for Lowland and Coastal Area, Universitas Pembangunan Nasional Veteran Jawa Timur, Surabaya 60294, Indonesia.

*Corresponding author: dedin.tp@upnjatim.ac.id

ABSTRACT

Cookies are a type of biscuit made from soft dough, characterized by high fat content, crispness, and a less compact cross-section when broken. Local foods such as black rice and black soybeans can be processed into antioxidant-rich flours, serving as substitutes for wheat flour. These ingredients contain antioxidants that help prevent the risk of chronic diseases. This study aimed to determine the effects of the proportion of black rice flour to black soybean flour and the addition of sunflower seed oil on the antioxidant activity of cookies. A factorial completely randomized design (CRD) with two factors. The First factor was the proportion of black rice flour to black soybean flour at three levels: 65%:35%, 75%:25%, and 85%:15%. The second factor was the addition of sunflower seed oil at three levels: 35%, 45%, and 55%. Data were analyzed using ANOVA at a 5% significance level, followed by Duncan's Multiple Range Test (DMRT) if significant differences were found. The results showed that the highest antioxidant was a proportion of black rice flour to black soybean flour of 65%:35% with 55% sunflower seed oil addition. This treatment resulted in cookies with 58.24 mg GAE/g total phenolic content, 86.64% antioxidant activity, and sensory scores of 4.64 (like) for color, 4.88 (like) for aroma, 4.76 (like) for taste, and 3.16 (quite like) for texture.

Keywords: cookies, black rice flour, black soybean flour, sunflower seed oil.

INTRODUCTION

Cookies are a form of food product preparation made from flour, sugar, and fat. Cookies are known by many people, both children, teenagers and adults, who live in rural and urban areas (Hariadi et al., 2022). The characteristics of good cookies are cookies that have a brownish yellow color or according to the color of the ingredients, a crunchy texture (brittle), aroma that is produced according to the ingredients used, the sweet taste of the sugar used and the characteristics of the ingredients used. Factors that influence the characteristics of cookies themselves are the formula of ingredients and additional ingredients must be balanced, the duration of mixing the dough, and the duration of baking cookies (Indrial, 2015). Black rice contains bioactive compounds such as anthocyanins 52.4 - 126.1 mg / 100 grams, total phenolic compound content 261.7 - 353.0 mg GAE / 100 grams which act as antioxidants, anti-inflammatories and have other important benefits for health. Antioxidant compounds in pigmented rice play a major role in preventing and reducing the risk of non-communicable diseases such as cardiovascular, hyperlipidemia, hyperglycemia and for the prevention of various types of cancer and other chronic diseases. Black rice has a high antioxidant and anthocyanin content. Anthocyanin is an organic compound from the flavonoid group. The anthocyanin pigment is found in the aleurone layer of black rice. The darker the color of the rice indicates the higher the anthocyanin content (Pang, 2018). Black soybeans have a protein content of 40.4g/100g, and a total polyphenol of 6.13 mg/g, flavonoids 2.19 mg/g, and anthocyanins 11.58 - 20.18 mg/g which is higher than yellow soybeans (Malencic 2012). The benefits of black soybeans themselves can reduce the risk of stroke, prevent premature aging, and as a source of antioxidants. The advantages of black soybeans are that they contain higher anthocyanins and have a longer shelf life compared to yellow soybeans (Ruslianti, 2017). Black soybeans have their own advantages because of their fairly high nutritional content, especially protein and carbohydrates.

The average soybean protein content is 35% and has a more complete amino acid composition than other nuts (Koswara 2009). Sunflower seed oil has a higher nutritional quality than other vegetable oils.

Sunflower seeds have quite high antioxidants. Sunflower seeds that have been processed into oil have an IC₅₀ value of 88.372 µg/mL which is included in the category of strong antioxidant activity, because it is still in the range of 50-100 µg/mL (Susanti et al., 2020). The composition of sunflower seed oil ranges from 23-45%. Sunflower seed oil contains 44-72% linoleic acid and 19% oleic acid. Sunflower seed oil is used for various purposes such as cooking oil, making margarine, cosmetic raw materials, and medicines, in addition, the cake or dregs from oil pressing contain 13-20% protein, which can be used as animal feed. For example, sunflower seeds are included in the low-cholesterol oil group, competing with corn oil, peanut oil and soybean oil, so they are very good for health (Rukmana, 2018). This study aims to determine the combination of black rice flour and black soybean flour with the addition of sunflower seed oil in high-antioxidant cookies.

MATERIAL AND METHODS

Materials

The materials used in this study were black rice (*Oryza sativa L.*) flour, black soybean (*Glycine soja L. Merrit*) flour, and ganyong (*Canna discolor L. syn.*) flour obtained from online stores, tapioca flour and sunflower seed oil obtained from Surabaya Supermarket. Other materials used were lecithin, ganyong flour, tapioca flour, vanilla, egg white, baking powder, coconut sugar, salt, skim milk.

Methodology

This study used a Completely Randomized Design (CRD) factorial pattern, consisting of two factors with two replications. Factor I is the proportion of substitution of black rice flour and black soybean flour (65%: 35%, 75%: 25%, 85%: 10%). Factor II is the addition of sunflower seed oil (35%, 45%, 55%). Data from the analysis results were processed using Analysis of Variance (ANOVA) with a level of 5%. If there is a significant difference, further testing is carried out using the DMRT (Duncan Multiple Range Test) 5% method. Data analysis used Microsoft Excel and SPSS 25 for Windows. The stages of making cookies are by doing the first mixing consisting of sunflower seed oil (35%, 45%, 55%), 1 gram of lecithin, and 20 grams of egg white. Then the second mixing is to add black rice flour: black soybean flour (65%: 35%, 75%: 25%, 85%: 15%), 1 g of salt, 40 g of coconut sugar, 1 of baking powder, 0.5 g of vanilla, 20 of skim milk, 30 g of tapioca flour, and 20 g of Ganyong flour. If it is homogeneous, the dough was moulded and the cookies were baked at 120°C for 35 min. The analysis carried out is Total Phenol Analysis, Folin-Ciocalteu Method, Antioxidant Activity Analysis, DPPH Method, and Organoleptic Hedonic Method including color, aroma, taste, and texture.

RESULTS AND DISCUSSION

1. Total Phenol

The average value of total phenol in cookies with the treatment of the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil can be seen in Table 1.

Table 1. The average value of total phenol cookies with the treatment of the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil.

Proportion of Black Rice Flour : Black Soybean Flour	Total Phenol (mgGAE/gr)		
	Sunflower Seed Oil 35%	Sunflower Seed Oil 45%	Sunflower Seed Oil 55%
65% : 35%	50.81 ^f ± 0.11	55.38 ^g ± 0.01	58.24 ^h ± 0.01
75% : 25%	44.35 ^c ± 0.26	46.18 ^d ± 0.04	48.78 ^e ± 0.16
85% : 15%	36.60 ^a ± 0.30	38.68 ^a ± 0.25	41.39 ^b ± 0.34

The proportion of black rice flour and black soybean flour 85%:15% with 35% sunflower seed oil produced the lowest total phenol (36.60 mgGAE/gr). In the treatment of 65%:35% with the addition of 55% sunflower seed oil produced the highest total phenol (58.24 mgGAE/gr). This is because black rice flour has a lower total phenol (9.49 mgGAE/gr) compared to black soybean flour (47.58 mgGAE/gr) and sunflower seed oil contains phenol which causes an increase in the total phenol of cookies. Kreps (2014) stated that sunflower seed oil contains phenol around 10-42 mg/g. The total phenol results are expressed as milligrams of gallic acid equivalent (GAE)/gram. Gallic acid is a natural phenolic compound found in several plants with health effects. Gallic acid is commonly used as a standard in measuring total phenol with Folin-Ciocalteu reagent because it is one of the naturally occurring phenols, stable and relatively inexpensive compared to other phenols (Kahkeshani et al., 2019). The graph of the effect of the proportion of black rice flour and black soybean flour with the addition of sunflower seed oil can be seen in Figure 1

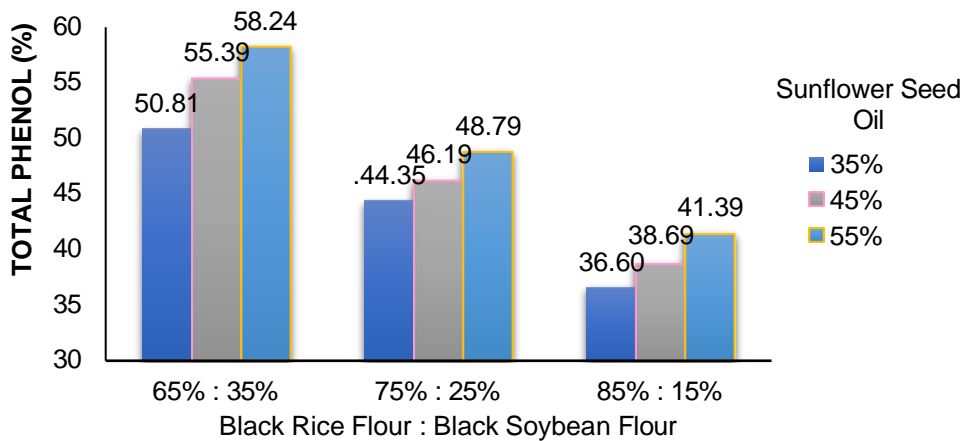


Figure 1. Relationship between the treatment of the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil to the total phenol of cookies.

2. Antioxidant Activity

The average value of antioxidant activity in cookies with the treatment of the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil can be seen in Table 2.

Table 2. Average value of antioxidant activity of cookies with the treatment of the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil.

Proportion of Black Rice Flour and Black Soybean Flour	Antioxidant Activity		
	Sunflower Seed Oil 35%	Sunflower Seed Oil 45%	Sunflower Seed Oil 55%
65% : 35%	76.10 ^a ± 0.87	81.18 ^c ± 0.04	86.64 ^f ± 0.048
75% : 25%	68.04 ^b ± 0.20	73.97 ^d ± 0.00	82.77 ^g ± 0.82
85% : 15%	60.27 ^e ± 0.09	71.27 ^f ± 0.04	81.33 ^h ± 0.05

The treatment of the proportion of black rice flour and black soybean flour 65%:35% with the addition of 55% sunflower seed oil produced the highest antioxidant activity (86.64%), while the treatment of 85%:15% with the addition of 35% sunflower seed oil produced the lowest antioxidant activity (60.27%). This is because black rice flour has lower antioxidant activity (32.53%) than black soybean flour (56.16%) and sunflower seed oil has high antioxidant activity. Susanti (2020) state sunflower seed oil has an antioxidant activity of 88.372 µg/mL. The graph of the effect of the proportion of black rice flour and black soybean flour with the addition of sunflower seed oil can be seen in Figure 2.

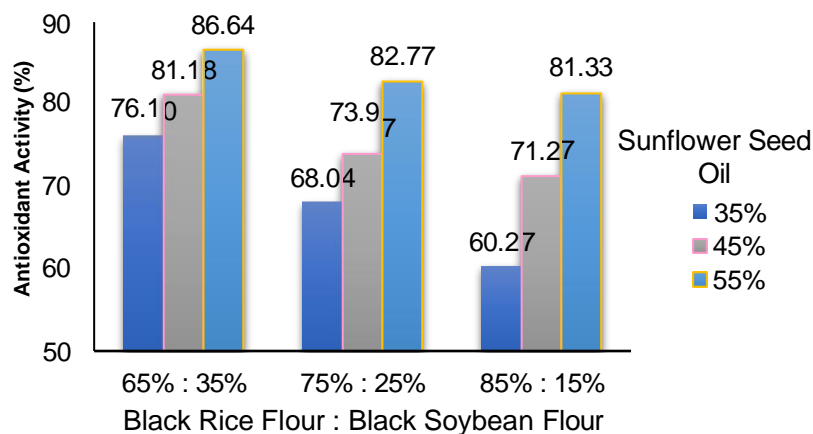


Figure 2. Relationship between the proportion of black rice flour and black soybean flour and the addition of sunflower seed oil on the antioxidant activity of cookies.

3. The organoleptic test results of cookies

The highest value was found in cookies with the treatment of the proportion of black rice flour and black soybean flour (65%:35%) and the addition of 35% sunflower seed oil which gets a average value of color scoring of 5.12 (very like). The smallest value was found in cookies with the treatment of the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 45% sunflower seed oil which produces an average scoring value of 4.12 (like). The addition of black rice flour was getting less and the addition of black soybean flour was getting more and more causes the color of the cookies to become browner. This is because black soybean flour has a protein containing free amino acids (lysine) which reacts with reducing sugars so that the resulting color is getting darker. According to Marco (2008) the addition of black soybean flour can also increase lysine and maltose which are involved in non-enzymatic browning reactions. The first stage of the Maillard reaction is the formation of Schiff bases which come from the reaction between the carbonyl group in reducing sugars with free amino acids, peptides or proteins (Rosida et al., 2006).

Table 3. Average of Total ranking of organoleptic test of cookies.

treatment			average		
The proportion of black rice flour and black soybean flour	sunflower seed oil	Colour	Aroma	Taste	Texture
65% : 35%	35%	5.12	4.92	4.56	3.12
	45%	4.64	4.92	4.88	3.64
	55%	4.64	4.88	4.76	3.16
75% : 25%	35%	5.04	4.92	5.08	5.80
	45%	4.12	4.40	4.40	2.96
	55%	4.16	4.32	4.12	2.88
85% : 15%	35%	5.08	4.80	4.92	4.96
	45%	4.56	4.80	4.88	4.12
	55%	4.88	5.12	4.96	4.68

The highest average aroma scoring value (5.12 very like) was found in cookies with the proportion of black rice flour and black soybean flour (85%:15%) and the addition of 55% sunflower seed oil. The smallest average aroma scoring value (4.40 liked) was found in cookies with the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 45% sunflower seed oil. Aroma is one of the main factors that consumers pay attention to when trying a product. The aroma produced in general from these cookies is the distinctive aroma of black rice flour and black soybean flour. The aroma produced can come from the flour during the cooking process. Susilawati et al., (2018) state during the cooking process, some of the contents in the flour undergo a gelatinization process so that the aroma of the flour produced affects the aroma of the resulting product. The highest value of the average taste score (5.08 very like) was found in cookies with the treatment of the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 35% sunflower seed oil. The smallest value of the average taste score (4.12 like) was found in cookies with the treatment of the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 25% sunflower seed oil. The taste of a food ingredient can come from the food ingredient itself and if it has undergone treatment and processing, then the taste is influenced by the ingredients added during the processing process.

The highest value of the average texture preference score (5.80 very much like) is found in cookies with the treatment of the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 35% sunflower seed oil. The smallest value of the average texture scoring value (2.88 dislike) is found in cookies with the treatment of the proportion of black rice flour and black soybean flour (75%:25%) and the addition of 55% sunflower seed oil. This is because the fat content of the cookies affects the texture or hardness of the cookies. The fat content of the raw materials shows that black rice flour is lower (3.19%) than black soybean flour (20.65%). The fat content functions to provide a soft texture to the cookies.

The results of Sadrina et al's research (2024) obtained the best cookie treatment from the proportion of black rice flour and black soybean flour (85%:15%) with the addition of 55% sunflower seed oil. This treatment produced a water content of 6.54%, ash content of 1.02%, fat content of 12.41%, protein content of 13.97%, and carbohydrate content of 57.31%. Furthermore, anthocyanin content test was carried out which produced $40.33\% \pm 0.106$ ppm. Anthocyanin is a natural free radical scavenger or better known as a natural antioxidant compound (Barrowclough, 2015). The test results in this study were higher than the literature of Kristanti (2020) which stated that the anthocyanin content of biscuits made from black rice flour with mung bean flour substitution was 36.98 ppm. The anthocyanin results obtained were influenced by the raw materials. Black rice flour has an anthocyanin content of 14.14 (Latifa, 2019). The black color of black rice is caused by the anthocyanin pigment found in the outer layer (Suardi, 2009). Black soybean flour has an anthocyanin content

of 20.18% (Adie, 2018). The black color of its skin causes black soybeans to have a higher anthocyanin content of 29 ± 0.56 mg/g compared to yellow soybeans 0.45 ± 0.02 mg/g (Agustina, 2016). Anthocyanins function as antioxidants to stop the reaction of free radical formation in the blood (Agustina, 2016).

CONCLUSION

The proportion of black soybean flour and sunflower seed oil increased, resulting in greater antioxidant content of cookies. This optimal cookie formula was obtained from the treatment of the proportion of black rice flour and black soybean flour (65%:35%) with the addition of 55% sunflower seed oil. This formula has a total phenol content of 58.24 mgGAE/gr, antioxidant activity of 86.64% and organoleptic values of color 4.64 (like), aroma 4.88 (like), taste 4.76 (like), texture 3.16 (like).

REFERENCES

- Adie, M. M., & Krisnawati, A. (2018). Kedelai Hitam: Varietas, Kandungan Gizi Dan Prospek Bahan Baku Industri. Badan Litbang Pertanian, Balai Penelitian Tanaman Kacang-kacangan dan Umbian-umbian.
- Agustina, A. W., & Anjani, G. (2016). Cookies tepung beras hitam dan kedelai hitam sebagai alternatif makanan selingan indeks glikemik rendah (Doctoral dissertation, Diponegoro University).
- Barrowclough, R. A. (2015). The effect of berry consumption on cancer risk. *Journal of Nutritional Health & Food Engineering*, 2(1), 1-9.
- Hariadi, H., Wibawa, I., Rahmawati, L., dan Riana, A. 2022. Pengaruh penambahan ekstrak jahe merah terhadap karakteristik organoleptik dan kandungan antioksidan cookies labu kuning. In *Prosiding Seminar Nasional* (pp. 335-344).
- Hartono, S.A.N., Rosida, D.F., Anggreini, R.A. 2024. Kajian Nutrisi Cookies Beras Hitam (*Oryza Sativa* L) da Kedelai Hitam (*Glycine Soja* L. Merrit). *Indonesian Journal of Microbiology* Vol: 1, No 4, 2025, Page: 1-12
- Indrial. 2015. Cookies Tepung Garut (*Maranta arudinaceace* L.) dengan Pengkayaan Serat pangan. Skripsi. Universitas Gadjah Mada. Yogyakarta.
- Kahkeshani., Zhang, X. 2019. Effect of Extraction and Drying Methods on Antioxidant Activity. *Food Bioprod. Process.* 99(13) 197-203
- Kristianti. 2020. Eksperimen Pembuatan Biskuit Tepung Beras Hitam Substitusi Tepung Kacang Hijau. *Jurnal Pangan dan Gizi*. 8. 13-21.
- Koswara, S. 2009. Seri Teknologi Pangan Populer (Teori Praktek). Teknologi Pengolahan Roti. e-BookPangan.com.
- Kreps, F., Vrbiková, L., & Schmidt, Š. (2014). Industrial rapeseed and sunflower meal as source of antioxidants. *Int J Eng Res Appl*, 4, 45-54.
- Adie, M. M., & Krisnawati, A. (2018). Black Soybeans: Varieties, Nutritional Content and Prospects for Industrial Raw Materials. Agricultural Research and Development Agency, Research Institute for Legumes and Tubers.
- Agustina, A. W., & Anjani, G. (2016). Black rice flour and black soybean cookies as an alternative low glycemic index snack (Doctoral dissertation, Diponegoro University).
- Barrowclough, R. A. (2015). The effect of berry consumption on cancer risk. *Journal of Nutritional Health & Food Engineering*, 2(1), 1-9.
- Hariadi, H., Wibawa, I., Rahmawati, L., and Riana, A. 2022. The effect of adding red ginger extract on organoleptic characteristics and antioxidant content of yellow pumpkin cookies. In *Proceedings of the National Seminar* (pp. 335-344).
- Hartono, S.A.N., Rosida, D.F., Anggreini, R.A. 2024. Nutritional Study of Black Rice Cookies (*Oryza Sativa* L) and Black Soybeans (*Glycine Soja* L. Merrit). *Indonesian Journal of Microbiology* Vol: 1, No 4, 2025, Page: 1-12
- Indrial. 2015. Arrowroot Flour Cookies (*Maranta arudinaceace* L.) with Dietary Fiber Enrichment. Thesis. Universitas Gadjah Mada. Yogyakarta.
- Kahkeshani., Zhang, X. 2019. Effect of Extraction and Drying Methods on Antioxidant Activity. *Food Bioprod. Process.* 99(13) 197-203
- Kristianti. 2020. Experiment of Making Black Rice Flour Biscuits Substituting Mung Bean Flour. *Journal of Food and Nutrition*. 8. 13-21.
- Koswara, S. 2009. Popular Food Technology Series (Theory and Practice). Bread Processing Technology. e-BookPangan.com.
- Kreps, F., Vrbiková, L., & Schmidt, Š. (2014). Industrial rapeseed and sunflower meal as source of antioxidants. *Int J Eng Res Appl*, 4, 45-54.
- Latifa, N., Nurhidajah, M. Y., & Yusuf, M. (2019). Anthocyanin stability and antioxidant activity of black rice flour based on packaging type and storage time. *Journal of Food and Nutrition*, 9, 27-40.
- Malencic, D., Cvejic, J., and Milandinovic, J. 2012. Polyphenol Content and Antioxidant Properties of Colored Soybean Seeds from Central Europe. *Journal of Medicinal Food*, 15.
- Marco. 2008. Technology of Biscuits, Crackers, and Cookies. Cambridge: Woodhead Publishing Limited.
- Pang, Y., Ahmed, S., Xu, Y., Beta, T., Zhu, Z., Shao, Y., and Bao, J. (2018). Bound phenolic compounds and antioxidant properties of whole grain and bran of white, red and black rice. *Food Chemistry*, 240 (January 2017), 212–221. <https://doi.org/10.1016/j.foodchem.2017.07.095>
- Rosida, D.F., Fardiaz, D., Apriyantono, A., Andarwulan, N. 2006. Isolation and Characterization of Soy Sauce Melanoidin and its Role as Antioxidant. Vol. 17 No. 3 (2006): *Journal of Food Technology and Industry*. p. 204-213
- Rukmana. R. 2000. Garut: Cultivation and Post-Harvest. Kanisius. Yogyakarta.
- Ruslianti. 2017. Applied Nutrition. Bandung. PT Remaja Rosdakarya Offset.
- Suardi, D. and I. Ridwan. 2009. Black rice, nutritious food that is not yet popular. *Agricultural Research and Development News* 31(2): 9-10.

- Susanti, Y., purba, A. V., and Rahmat, D. 2020. Antioxidant Value and SPF of Combination of Sesame Seed Oil (*Sesamum indicum* L.) and Sunflower Seed Oil (*Helianthus annuus* L.). *Pharmaceutical Journal*. 16(1): 107-110.
- Susilawati, S., Sugiharto, R., & Damaiyanti, S. M. (2018). Formulation of Virgin Coconut Oil (VCO) and Soy Lecithin Emulsifier on Emulsion Stability and Sensory Characteristic of Red Beans Paste. *Journal of Agricultural Product Technology & Industry*, 21(1), 42-50.