EFFECTIVENESS OF FERMENTED BLACK GLUTINOUS RICE SNACK BAR INTERVENTION ON ADULT IMMUNITY STATUS (INTERLEUKIN-6)

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Abstract **Background**: Increased oxidative stress has a negative impact on several components of the cell membrane, namely damage to membrane lipids to form malonaldehyde. Free radicals can also cause the secretion of inflammatory mediators, namely IL-6 which acts as a pro-inflammatory cytokine. The content of Fermented Black Glutinous Rice Snack Bar is also expected to be able to reduce inflammation which is indicated by a decrease in IL-6 levels as an indicator of inflammation.
Objective: This study aimed to know effectiveness of fermented black glutinous rice (FBGR) snack bars on adult immunity status (IL-6).
Method: The study design was true experiment with a control group. There were 20 adult subjects (>20 years old) each group by purpose sampling method. Subjects were divided into two groups: the experimental group (FBGR snack bars administrated for 30 days) and the control group (no FBGR snack bars administration). IL-6 levels were collected pre and post intervention. Data analysis by double difference approach to compare immunity status (IL-6 levels alteration) of the two groups.
Results: Wilcoxon Sign Rank statistic test showed that was significant difference IL-6 level pre and post intervention in control group (95% CI); significant difference IL-6 level reduced pre and post intervention in intervention group (95% CI). Independent T test showed that significant difference IL-6 level between intervention and control groups with p<0,001 (p<0,05) in 95% CI.
Conclusion: This study concluded that FBGR snack bars intervention reduced IL-6 levels.

Keywords: FBGR snack bars, immunity status, IL-6 levels.

BACKGROUND

Vitamins and minerals are essential for maintaining an optimal immune system. Since most vitamins and minerals cannot be synthesized by the body, consuming a varied and balanced diet is essential for sources of vitamins and minerals such as fruits, vegetables and animal foods. Some vitamins and minerals act as antioxidants that greatly affect the quality of human life, containing vitamins A, E, C, selenium, iron and zinc. These nutrients are needed in the body's defense system because of their role as antioxidant nutrients (1)
Black sticky rice tape is one of the foods that contains anthocyanins and has antioxidant activity and fiber. Black sticky rice tape produced in West Bandung Regency is one of the foods that contains anthocyanins with anthocyanin content of black sticky rice tape of 3.48mg/100g, while the total phenol of black sticky rice tape is 73.38 mg/100gr. The antioxidant activity of black sticky rice tape is 70.2% (2). Ethanol content of black sticky rice tape is 1.14, total sugar content of black sticky rice tape is 18.31% and pH content of black sticky rice tape is 3.83 and total acid of black sticky rice tape is 0.91%. The crude fiber content of black sticky rice tape consumed by respondents in this study was 1.32% with dietary fiber content of 5.9% (3). In a previous study conducted by Fauziah Nur, 2015, it was shown that in West Bandung Regency, the amount of black sticky rice tape consumption of at least >11.5 grams per day can prevent the occurrence of metabolic syndrome, giving black sticky rice tape as much as 200 grams every day in West Java Province and in West Sumatra, was proven to have a significant effect on improving metabolic syndrome components and there was a significant improvement in other profiles, namely total cholesterol and LDL cholesterol levels and a decrease in waist circumference ($p < 0.001$) (4). The results of a 2017 study showed that giving steamed black sticky rice brownies was proven to have a significant effect on reducing waist circumference in people with abdominal obesity ($p<0.001$) (5) and giving black sticky rice tape Snack Bar was proven to have a significant effect on improving lipid profiles in people with dyslipidemia ($p<0.001$) (6) and giving black sticky rice tape snack bars was effective in improving blood glucose levels in prediabetic patients (6). Zeng defines oxidative stress as a state of imbalance between free radicals and antioxidants, where the number of free radicals is greater when compared to antioxidants. The production of free radicals exceeds the ability of intracellular antioxidants to neutralize them, so excess free radicals have the potential to cause cell damage. This damage is oxidative damage, namely damage to the biomolecules that make up cells caused by their reaction with free radicals. Increased oxidative stress has a negative impact on several components of the cell membrane, namely damage to membrane lipids to form malonaldehyde. Free radicals can also cause the secretion of inflammatory mediators, namely IL-6 which acts as a pro-inflammatory cytokine (7). The content of the Black Glutinous Rice Tape Snack Bar is also expected to be able to reduce inflammation which is indicated by a decrease in IL-6 levels as an indicator of inflammation. Based on the description above, it is necessary to conduct research on the Effectiveness of Giving Fermented Black Glutinous Rice Snack Bars on Immunity Status.

METHODS

S*tudy setting and design*

This study was an experimental design with a control group. In the intervention group, samples were given FBGR snack bar for 30 days, while in the control group were not given FBGR snack bar. The research was conducted in Puskesmas Sawah Lega Kabupaten Bandung West Java on October 2021.

Study population and sampling procedure

Study population was the employees of Puskesmas Sawah Lega Kabupaten Bandung, West Java. Samples were employees that met the criteria and agree to participate the research by signed informed consent. Minimum samples were calculating by hypothesis

test to compare two means. There were 40 samples totally or 20 subjects for each group. The sample criteria were female and male, aged >20 years old, willing to participate this research. Exclusion criteria were stroke or CAD or renal diseases sufferers, professional athlete, and not willing to participate the research. Samples were determined using purposive sampling technique.

Data Collection and Variable measurement

The data collection was performed using primary data by interviewing samples. Immunity status (IL-6 level) were collected before FBGR snack bars were given, and after 30 days intervention. Normality test for IL-6 data was done to check normality distribution data. Bivariate analysis to test efficacy the FBGR snack bar by independent and the dependent t-test when data normally distributed, Mann Whitney and Wilcoxon Sign Rank test when data not normally distributed.

Ethic Consideration

The participants provided their written informed consent to participate in this study.

RESULT AND DISCUSSION

FBGR Snack Bar Nutrient Content

FGBR snack bar is a snack that made form fermented black glutinous rice and black glutinous rice flour. The taste is sweet and acid unique from fermented black glutinous rice. Texture is compact and soft. The formulae FBGR snack bar consisted of 95% fermented black glutinous rice and 5% black glutinous rice flour. Form 200 g fermented black glutinous rice and 10 g black glutinous rice flour, produced six pieces snack bar with 30 g each.

Figure 1. FBGR snack bar and the packaging

Nutrients and energy content of snack bar were examined in Laboratorium Balai Besar Penelitian dan Pengembangan Pascapanen Pertanian, Kementerian Pertanian, Bogor. Tabel 1. Shows nutrients and energy content of snack bar.

Tabel 1. Nutrients and Energy Total Content of FBGR Snack Bar

Nutrient	Analysis Method	Per 100 g	Serving size 30 g	Percent Dietary Allowance Snack (8)
Karbohidrat	By Different	50,41 g	15,12 g	86,6%
Protein	Kjeldahl	10,97 g	3,29 g	84,2%
Fat	Soxhlet	2,91 g	0,87 g	58,9%
Energy Total	Calculation	271,71 kkal	81,51 kkal	81.20%
Anthocianin	Spectrophotometri	21,918 mg	6,5754 mg	65,75%
Fibre	Gravimetri enzymatic	5,8 g	1,74 g	69,60

Tabel 1 shows that FBGR snack bar is a healthy choice to maintain adult weight because energy content low than the RDA , low in fat and carbohydrate but good source in anthocyanin and fiber. Maheshwari et.al stated to maintain a robust immune system, it is necessary to consume a well-balanced diet that provides all of the necessary nutrients in appropriate quantities (9).

IL-6 level of Intervention and Control Group
The IL-6 levels were examined pre and post intervention in intervention and control group. Table 2, 3 and 4 shows data for IL-6.

Table 2. Comparison of Pre-Post IL-6 levels in Control Group

Variables	Control group				Nilai p*)
	pre		post		
	mean	SD	mean	SD	
IL-6 level (pg/mL)	2,95	1,49	5,46	1,86	<0,001

*) *Wilcoxon Sign Rank Test*

Wilcoxon Sign Rank statistic test showed that was significant difference IL-6 level rose pre and post intervention in control group (95% confidence interval).

Table 3. Comparison of Pre-Post IL-6 levels in Intervention Group

Variables	Intervention group				Nilai p*)
	pre		post		
	mean	SD	mean	SD	
IL-6 level (pg/mL)	3,48	1,57	3,37	1,43	<0,001

*) *Wilcoxon Sign Rank Test*

Wilcoxon Sign Rank statistic test showed that was significant difference IL-6 level reduced pre and post intervention in intervention group (95% confidence interval).

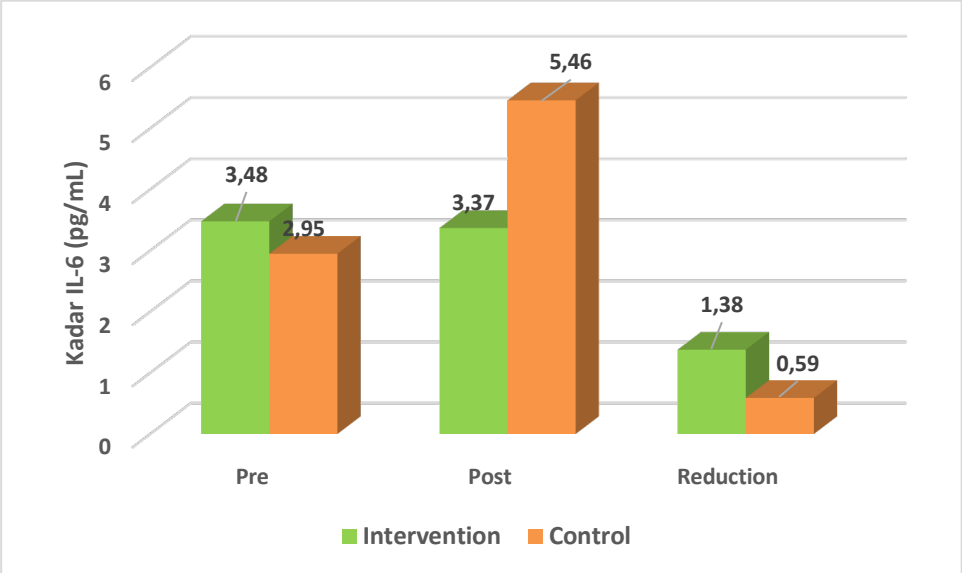
Tabel 4. IL-6 levels Comparison between Intervention and Control Groups

Variables	Intervention		Control		Nilai P
	Mean	SD	Mean	SD	
IL-6 (pg/mL) (pre)	3,48	1,57	2,95	1,49	0,123
IL-6 (pg/mL) (post)	3,37	1,43	5,46	1,86	<0,001
Reduction IL-6 (pg/mL)	1,38	0,895	0,59	0,315	<0,001

*)*Independent T Test*

Table 4 shows that independent T test showed that significant difference IL-6 level between intervention and control groups with p<0,001 (p<0,05) in 95% confidence interval. Therefor it can be concluded that there is an effect of FBGR snack bar intervention to immune status based on reduction of IL-6 level. This result in line with some studies. Liu J. et al. founded that anthocyanins have pharmacological potential for diseases of the circulatory, endocrine, digestive, urinary, sensory, nervous and immune systems (10). Li et al added that anthocyanins can activate the immune system during infection by increasing the release of cytokines and activating immune cells (11). This natural products can modulate the immune

system by enhancing or suppressing the immune response (12). Anthocyanin is often better tolerated than conventional pharmaceutical treatments, making this an attractive supplement for enhancing immune system function (9). Kozłowska and Dzierżanowski stated anthocyanin have been proven to play important role in regulating inflammatory pathway of proinflammatory cytokines, one of them is IL-6 (13).



Graph 1. IL-6 levels Comparison between Intervention and Control Groups

CONCLUSION

There is an effect of giving fermented black glutinous rice snack bars on immune status based on reduction of IL-6 level. Further research is need to conduct.

COMPETING INTERESTS

All authors had none to declare

AUTHOR'S CONTRIBUTION

Yenny Moviana conceived of the presented idea, data analysis, and writing manuscript; Rr. Nur Fauziyah and Wida Purbaningsih were in charge of data collection and analysis; and drafting the manuscript. All authors contributed to the final manuscript.

REFERENCES

1. Siswanto, Budisetyawati, & Ernawati, F. (2013). Peran Beberapa Zat Mikro Dalam Sistem Imunitas. *Gizi Indon*, 57-64.
2. Trinovani, Elvi, Afifah Riska Rafa, Fauziyah, Roro Nur. 2020. Determination of Antosianin Total Levels and Antioksidant Activities in Black Glutinous Rice Extract and Fermented Black Glutinous Extract. International Medical Journal. Vol 25, Issue 05. Artikel https://www.seronijihou.com/volume/I MJ/25/5/determination-of-antosianintotal-levels-and-antioxidant-activitiesin black-glutinous-rice-extract-andfermented-black-glutinous-rice-extract5ec8099a8eb18.pdf
3. Fauziyah, Roro Nur, Mimin AMinah, Osman Syarief, Holil M Par'I, Widi Hastuti, Surmita. 2020. Effectiveness of Steamed Brownies Base on Fermented Black

GlutinousRice on Decreased Waist Circumference in Abdominal Obesity. Jurnal Ilmu dan Teknologi Kesehatan. Vol x No x. Artikel http://ejurnal.poltekkesjakarta3.ac.id/index.php/jitek

4. Fauziyah, Roro Nur, Fitriani Nurjannah, Surmita. 2020. Effect Of Fermented Glutinous Black Rice on LDL Cholesterol Level. Jurnal Riset Kesehatan. Vol 12 No.1 Tahun 2020. Artikel https://juriskes.com/index.php/jrk/article/view/1759
5. Fauziyah, Roro Nur, Fitriani Nurjannah, Surmita. 2020. Effect Of Fermented Glutinous Black Rice on LDL Cholesterol Level. Jurnal Riset Kesehatan. Vol 12 No.1 Tahun 2020. Artikel https://juriskes.com/index.php/jrk/article/view/1759
6. Zheng W. and Wang S.Y., 2009. Antioxidant Activity and Phenolic Compounds in Selected Herbs. J.Agric.Food Chem., 49 (11) : 5165-70, ACS Publications, WashingtonD.C.
7. Fauziyah, Roro Nur, Fitriani Nurjannah, Surmita. 2020. Effect Of Fermented Glutinous Black Rice on LDL Cholesterol Level. Jurnal Riset Kesehatan. Vol 12 No.1 Tahun 2020. Artikel https://juriskes.com/index.php/jrk/article/view/1759
8. Peraturan Menteri Kesehatan RI Nomor 28 tahun 2019 tentang Angka Kecukupan Gizi yang Dianjurkan untuk Masyarakat Indonesia.
9. Maheshwari, S.; Kumar, V.; Bhadauria, G.; Mishra, A. Immunomodulatory potential of phytochemicals and other bioactive compounds of fruits: A Review. Food Front. **2022**, 3, 221–238.
10. Liu J, Zhou H, Song L, Yang Z, Qiu M, Wang J, Shi S. Anthocyanins: Promising Natural Products with Diverse Pharmacological Activities. Molecules. 2021 Jun 22;26(13):3807. doi: 10.3390/molecules26133807. PMID: 34206588; PMCID: PMC8270296.
11. Li, B., Wang, L., Bai, W., Chen, W., Chen, F., Shu, C. (2021). Immune System Promotion and Antibacterial and Antivirus Effects of Anthocyanins. In: Anthocyanins. Springer, Singapore. https://doi.org/10.1007/978-981-16-7055-8_20
12. Ijinu TP, De Lellis LF, Shanmugarama S, Pérez-Gregorio R, Sasikumar P, Ullah H, Buccato DG, Di Minno A, Baldi A, Daglia M. Anthocyanins as Immunomodulatory Dietary Supplements: A Nutraceutical Perspective and Micro-/Nano-Strategies for Enhanced Bioavailability. Nutrients. 2023 Sep 26;15(19):4152. doi: 10.3390/nu15194152. PMID: 37836436; PMCID: PMC10574533.
13. Kozłowska A, Dzierżanowski T. Targeting Inflammation by Anthocyanins as the Novel Therapeutic Potential for Chronic Diseases: An Update. Molecules. 2021 Jul 20;26(14):4380. doi: 10.3390/molecules26144380. PMID: 34299655; PMCID: PMC8304181.