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The Amino Acid and Mineral Content of Black Oncom Processed with Fermentation Modifications

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ABSTRACT

The aim of this study was to analyze the effect of the starter and storage conditions of black oncom production and processing on the amino acid and mineral compositions. This research was conducted from June to December 2019 at IPB University utilizing a completely randomized design with treatments of the use of a *Rhizopus oligosporus* starter and different storage conditions. Amino acids assay were measured by UPLC (Ultra Performance Liquid Chromatography), while minerals were measured by AAS (Atomic Absorption spectrophotometer). Both amino acids and minerals were determined by the AOAC method. Black oncom was produced through a controlled fermentation process which had significantly higher amino acids ($p<0.05$) compared to black oncom made traditionally by traditional producers. In both controlled and traditional fermentation, the highest amino acid was glutamic acid (6.31±0.88 g/100 g; 3.85±0.62 g/100 g), while the lowest amino acid was methionine (0.003 g/100 g; 0.17±0.09 g/100 g), respectively, in dry basis. Likewise, the mineral content of calcium (189.54±32.69 mg/100 g) and zinc (9.49±0.77 mg/100 g) was also significantly higher in the controlled fermentation than in the traditional fermentation. In conclusion, the controlled production process of black oncom can produce higher amino acids and mineral content compared to using the natural fermentation process.

Keywords: amino acids, black oncom, controlled fermentation, minerals, traditional fermentation.

INTRODUCTION

Black oncom is a traditional food of the people of West Java made from peanut meal and fermented by *Rhizopus oligosporus* and is used as a source of protein (Kumbhare 2014). The process of peanut meal fermentation carried out by the people of West Java, especially the Bogor area, still utilizes the traditional natural fermentation process without the use of an additional yeast starter such as *Rhizopus oligosporus*, therefore making the natural fermentation process to be less effective, especially in increasing the digestibility of proteins (Elyas *et al.* 2002; Pranoto *et al.* 2013). Low protein digestibility will affect the components of the amino acid and mineral content (Joye 2019; Manditsera *et al.* 2019), while the controlled fermentation process is able to break the bonds of the amino acids and minerals that are bound with antinutrients to free them (Nkhata *et al.* 2018).

In addition to the natural fermentation process, black oncom craftsmen also use boxes made of bamboo as storage containers for the fermentation process which are then covered with sacks. The storage conditions of the fermentation process can improve the quality of a product (Cabello-Olmo *et al.* 2020; Han *et al.* 2003). This shows the need for conditions where the fermentation process can result in better quality black oncom using more modern equipment, such as plastic boxes. Therefore, a production process modification is needed in the fermentation process of black oncom using the standardized yeast starter of *Rhizopus oligosporus* and the use of a more suitable storage container for fermentation. The use of a *Rhizopus oligosporus* starter can improve the quality of black oncom, especially its protein content (Sastraatmadja *et al.* 2002; Basoni *et al.* 2019) which can also decrease aflatoxin (Kusumaningtyas *et al.* 2019; Ginting *et al.* 2019). According to these studies, the production process modification of black oncom is expected to be able to increase the components of amino acids and minerals. Therefore, the aim of this study was to describe

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the effect of the starter and storage conditions of black oncom production processing on the amino acid and mineral compositions.

METHODS

Design, location, and time

The design of this study was a completely randomized design with treatments of a *Rhizopus oligosporus* starter and different storage conditions. This research was conducted from June to December 2019 and the sample was analyzed duplicately at the Food Processing Laboratory, Nutrition and Food Analysis Laboratory, Department of Community Nutrition, and Chemistry Integrated Laboratory, IPB University, Bogor, West Java, Indonesia.

Materials and tools

The main ingredient of peanut meal was obtained from a peanut oil distributor in Sukasari, Bogor and natural fermented black oncom was obtained from a black oncom producer at Bantar Kambing, Bogor. The materials for black oncom production were water and the starter of *Rhizopus oligosporus*. The materials for analysis were HCl 6 N, ash-free filter paper No.42, internal standard of AABA, Accq. Tag Fluor Borrate Buffer, Accq. Tag reagent 2A, nitric acid, sulfuric acid, aquabidest and aquadest. Meanwhile, the tools of this research were a plastic box, bowl, spoon, stove, boiler, digital scale, glassware, headspace vial, syringe, UPLC and AAS.

Procedures

The production process of black oncom. The production process of black oncom is shown in Figure 1, where the differences between the production process in the laboratory and the craftsmen lie in the use of *Rhizopus oligosporus* starter and the storage container for the fermentation process. Fungi treatment was carried out to see differences in the quality of black oncom because the culture of *Rhizopus oligosporus* has better quality (Sastraatmadja *et al.* 2002; Feng *et al.* 2007). This was to investigate whether the natural fermentation process was less than optimal compared to the fermentation using a yeast starter.

Amino acid assay. A 0.1–1.0 g sample was weighed and placed into a 20 ml headspace vial, then 5 mL of HCl 6 N was added and the vial closed. The solution was heated in an oven at 110ºC for 22 hours. After heating, the vial containing the solution was cooled and the sample was transferred to a 50 ml volumetric flask. The headspace tube was rinsed and diluted with aquabides to the boundary mark and then homogenized. The sample solution was filtered with ash-free filter paper No.42, and then filtered once again with a 0.2 µm GHP/RC syringe filter. 500 µl of the solution was then pipetted and put in a 2 ml tube. 40 µl internal AABA standard 2.5 Mm and 460 µl aquabides were added into the sample solution, and then vortexed. The solution was derivatized by piping 10 µl of standard solution or sample that had been added by AABA internal standard into the insert vial. 70 µl Accq. Tag Fluor Borrate Buffer was added to the solution, and then vortexed. 20 µl Accq. Tag reagent 2A was again added to the solution, then vortexed. The solution was then heated in the heating block for exactly 10 minutes at 60ºC. The cooled solution was then injected into the UPLC system (AOAC 2005).

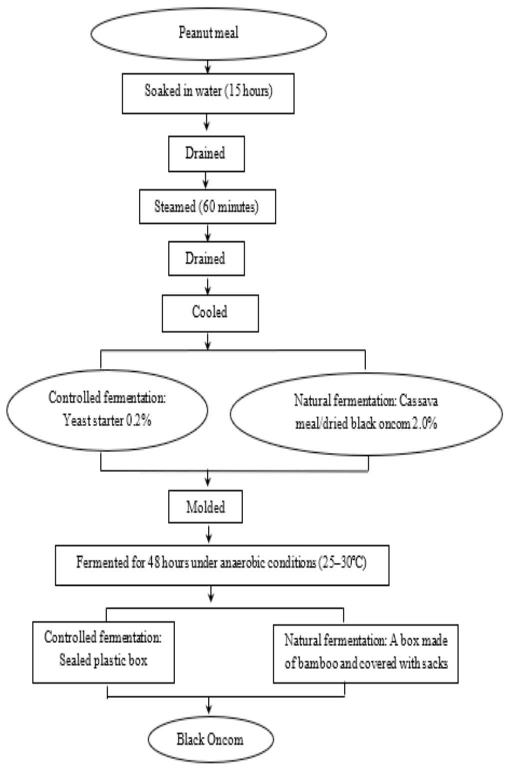

Figure 1. The production process of black oncom (modification from Sastraatmadja *et al.* 2002; Feng *et al.* 2007)

Mineral assay. Two (2) g of fine sample was put into 150 ml erlenmeyer and 5 ml of nitric acid and then concentrated sulfuric acid was added. The solution was heated on a hotplate until the white ash disappeared. The solution was then diluted with 100 ml aquades in a measuring flask and filtered using whatman filter paper. Standard solutions, blanks and samples were flowed to the AAS (Atomic Absorption Spectrophotometer) instrument. The absorbance of the standards, blanks, and samples were measured at the wavelength and parameters corresponding to each mineral (AOAC 2005).

Data analysis

Research data were processed using SPSS 16.0 for Windows. Independent t-test analysis was used to examine amino acid and mineral variables with p-values less than 0.05 ($p<0.05$) set as the significance limit.

RESULTS AND DISCUSSION

The results of the fermented production process of black oncom in controlled fermentation and natural fermentation can be seen in Figure 2. The research results showed that black oncom contained 16 amino acids, which are presented in Table 1. The results of the study showed that black oncom had a high amino acid content in the order of glutamic acid, aspartic acid, arginine, and leucine. The lowest amino acid content was in the form of the amino acid methionine. In addition, the production process modification of black oncom significantly affected amino acid content ($p<0.05$), where the black oncom made by the controlled process showed higher amino acid components compared to the black oncom made by craftsmen on all types of amino acids except for the amino acid methionine.

This showed that the production process of black oncom using a *Rhizopus oligosporus* starter and the treatment of storage for the fermentation process can produce better quality black oncom characterized by higher levels of amino acids in each component. These results are in line with the explanation from Pranoto *et al.* (2013) and Nkhata *et al.* (2018), where the controlled fermentation process can produce a final product that is better than the natural fermentation process because of the breakdown of amino acids that bind antinutrients into free amino acids.

On the other hand, the amino acid content of black oncom in this study was in accordance with the research results by Liu *et al.* (2012), where the amino acid component of fermented peanut meal in dry basis contained the maximum (8.72 g/100 g dry basis of glutamate) and the minimum (0.58 g/100 g dry basis of methionine). In addition, the amino acid content of black oncom in this study was in accordance with the study of Yang *et al.* (2016) that reports 6.19 aspartate, 1.30 threonine, 1.84 serine, 11.4 glutamate, 2.74 proline, 3.02 glycine, 2.56 alanine, 0.78 cysteine, 2.60 valine, 0.65 methionine, 1.93 isoleucine, 3.72 leucine, 2.22 tyrosine, 3.23 phenylalanine, 2.16 lysine, 1.44 histidine, and 4.07 arginine in g/100 g dry basis. Although there are differences with the results of this study, the highest amino acid content of black oncom is the same, i.e., glutamic acid, aspartic acid, arginine and leucine, respectively.

The high amino acid component in controlled black oncom showed that black

Figure 2. The fermented production process of black oncom (left) controlled fermentation (right) natural fermentation

Table 1. Amino acid contents of controlled black oncom and natural black oncom (g/100 g dry basis)

Amino acid	Controlled black oncom	Amino acids score*	Traditional black oncom	Amino acids score*
L-Lysine	1.40±0.27b	95	0.94±0.17[a]	72
L-Leucine	2.83±0.10[b]	100	1.65±0.25[a]	96
L-Isoleucine	1.67±0.06[b]	100	1.07±0.19[a]	100
L-Methionine	0.003 ±0[a]	23	0.17±0.09[a]	36
L-Phenylalanine	2.35±0.23[b]	100	1.18±0.05[a]	100
L-Tyrosine	1.14±0.10[b]	100	1.07±0.15[a]	100
L-Threonine	1.27±0.06[b]	100	0.76±0.08[a]	100
L-Valine	2.44±0.63[b]	100	1.25±0.09[a]	100
L-Histidine	1.07±0.12[b]	100	0.61±0.17[a]	100
L-Arginine	3.17±0.08[b]		1.49±0.46[a]	
L-Alanine	2.20±0.11[b]		1.31±0.10[a]	
L-Aspartic acid	3.79±0.53[b]		2.41±0.01[a]	
Glycine	2.43±0.12[b]		0.90±0.10[a]	
L-Proline	1.66±0.41[b]		n/a	
L-Glutamic acid	6.31±0.88[b]		3.85±0.62[a]	
L-Serine	1.83±0.08[b]		0.98±0.00[a]	

Data are represented in mean±SD; n/a: not analyzed; The different superscript (a and b) in one row mean significantly different ($p<0.05$); *: Report of a joint WHO/FAO/UNU expert consultation 2007

oncom can be used as a source of protein supplementation because the fermentation process can break down peanut meal protein into smaller peptide forms <24 kD (Liu *et al.* 2012), in which the formation of smaller peptides was the result of the fungi protease enzyme's action. This form of fermented peanut meal oligopeptide (<14 kDa) can further contribute to antioxidant activity (Zhang *et al.* 2014; Yang *et al.* 2016). The low molecular weight of amino acids (<1.4 kD) are very helpful in supporting human health in overcoming oxidative damage (Yang *et al.* 2016). The oxidative damage situation occurs in the elderly due to the aging process (Rimbach *et al.* 2005), therefore black oncom can be used as food that supports the health of the elderly.

However, black oncom still shows to have a limiting amino acid score; namely methionine and lysin in the controlled black oncom, while traditional black oncom is found to contain methionine, lysin and leusin. According to the amino acid score, the usage of amino acid in the body based on the lower amino acid score of controlled and traditional black oncom is only 23 or 36 respectively. Based on this knowledge, the application of black oncom for protein supplementation still requires other ingredients to maximize amino acid usage for the body.

Black oncom can also be utilized for its other nutrient content, i.e., mineral sources such as calcium, zinc and iron, which are described in Table 2. The calcium and zinc content of controlled black oncom was significantly higher ($p<0.05$) compared to traditional black oncom. This shows that the controlled production process of black oncom was able to produce high mineral components, especially calcium and zinc. The results of this study are in line with Nkhata *et al.* (2018), where a controlled fermentation process can increase the amount of minerals due to the breaking of bonds between minerals and antinutrient substances or fiber (Liang *et al.* 2008).

Table 2. Mineral content of controlled black oncom and natural black oncom (mg/100 g dry basis)

Mineral	Controlled black oncom	Natural black oncom
Calcium (Ca)	189.54±32.69[b]	123.55±6.32[a]
Zinc (Zn)	9.49±0.77[b]	3.29±0.23[a]
Iron (Fe)	2.54±1.49[a]	9.90±0.81[b]

data are represented in mean±SD; The different superscript (a and b) in one row mean significantly different ($p<0.05$)

An increased amount of calcium, zinc and iron minerals can be found in several fermented products in India such as sorghum flour and bambara nut (Pranoto *et al.* 2013). However, the iron content in traditional black oncom was significantly higher ($p<0.05$) compared to controlled black oncom. The low iron content of the controlled black oncom was probably due to the use of these minerals for the metabolic process of fungi, in which the minerals may be a nutrition source for fungi. While for the natural black oncom it was probably due to the use of cassava meal in the fermentation process. The mineral content of calcium and zinc in black oncom was greater than peanuts, while the iron content was smaller (Toomer 2018); the increase or decrease occurs due to the metabolic processes carried out by fungi in the black oncom. This shows that the controlled production process of black oncom can improve mineral content, especially calcium and zinc, therefore the use of black oncom as a fermentation product for health will be more optimal, especially in dealing with nutritional problems.

CONCLUSION

Black oncom produced through the controlled fermentation process had a higher amino acid content compared to black oncom derived from natural fermentation. Likewise, the mineral content of calcium and zinc in controlled black oncom was higher than that of traditional black oncom, while the iron content was lower. This revealed that the production process of black oncom in a controlled condition can improve the quantity of amino acids and some minerals, resulting in a product with better potential for health effects, especially in treating oxidative damage in the elderly.

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AUTHOR DISCLOSURES

All authors have no conflict of interest in the research.

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