# The Potential Applications of Modified Nagara Bean Flour through Fermentation for Innovation of High Protein Analog Rice

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***Abstract***--Rice is the staple food for most Indonesian people and the rate of rice consumption is increasing every year, eventhough government should go import. The source of carbohydrates not only from rice, but there are many other materials such as cereals, tubers that have not been threated optimally. Similarly, the occurrence of a shortage of protein in some communities in Indonesia are still being found. Therefore, the strategy of this problem is with food diversification to substitute or replace the rice needs by using rice analog from Nagara beans (*Vigna unguiculata* ssp *Cylindrica*). Nagara beans have high carbohydrate content about 50-60% and 20-25% protein. This research was aimed to modify Nagara bean flour through spontaneous fermentation with variation of bean size and periods of fermentation. Fermented Nagara bean flour were tested several parameters which include yield, moisture content, bulk density, protein content, water absorption capacity, swelling volume, and solubility. Nagara beans contain amino acid dominant aspartic acid 0.913%, glutamic acid 2.182% and histidine 0.826%. Grits size produce flour yield, protein content, water absorption capacity, and the swelling volume greater than others. Fermentation time up to 120 hours tend to lower flour yield and protein content, while the water absorption capacity tend to increase up to 96 hours fermentation and swelling volumer tends to be stable up to 96 hours fermentation periods.

*Keywords : nagara bean flour, spontaneous fermentation, protein, water absorption capacity*

## I. INTRODUCTION

One alternative in achieving national food security is the food diversification. But the culture of Indonesian people “it feels not full before eating rice” makes the process of diversification has not run smoothly. Therefore, we need an alternative food that resembles the staple food of Indonesia, namely rice. The food resembling rice is called analog rice. Analog rice generally processed from cereals or tubers with a dominant carbohydrate content, but on the other hand there are actually several types of legumes that contain carbohydrates dominant one of it which grow in wetland areas.

Nagara bean (*Vigna unguiculata* ssp. *Cylindrica*) is one of the local bean which grow in South Kalimantan, especially in Hulu Sungai Selatan area. The largest component in the Nagara bean is carbohydrate and implies almost the same as cowpea and green beans which ranges from 62%. On the other hand, Nagara bean is also one source of vegetable protein. Protein content ranged from 22.7 to 27% (Noor 1993), it is higher than green bean and cowpea.

According Niba (2003) limitations legume characteristics can be improved by fermentation and bioprocessing techniques. The fermentation process was known as one method that can modify starch structure and physicochemical properties of a material (Chinsamran *et al*. 2005), where the fermentation can affect the solubility, the development of granule, and viscosity of starch (Abia *et al*. 1993; Nche *et al*. 1994; Yadav and Khetarpaul 1994), the specific characteristics is very instrumental on subsequent product processing. Prinyawiwatkul *et al*. (1997) assess the functional properties of bean flour were influenced by soaking, boiling and fermenting fungi, and Yadav and Khetarpaul (1994) on *Phaseoulus mungo* fermentation process at a temperature of 25-30 ºC for 12 and 18 hours was able to increase the digestibility of starch from 57% to more than 88%.

The process of fermentation with soaking is a technique that is easy to modify starch, and on the other hand it is able to improve in vitro starch digestibility 17-23% after 12 hours of soaking. With longer soaking treatment, it is expected to increase digestibility of starch and contribute to the declining of antinutrition such as phytic acid and polyphenols which can inhibit the activity of α-amylase (Desphande and Cheryan 1984).

Material that has been fermented legumes are digested significantly faster than the legume is not fermented. This is possible because of the loss of structural integrity of the starch granules, changing the interaction between starch and fiber because inactivation of some antinutrien (such as phytic acid).

Natural fermentation process is simple and inexpensive method to reduce and eliminate oligosaccharides. Zamora and Fields (1979) states that fermented cowpea will reduce stachiosa and raffinose, this is caused by the ability of lactic acid bacteria to use oligosaccharides for metabolism. Lactic acid is the dominant role in the process of fermentation include *Lactobacillus casei*, *Lactobacillus leichmanni, Lactobacillus plantarum*, *Pediococcus pentosaceus and Pediococcus acidilactici.*

This study was aimed to assess the modification process Nagara bean flour through spontaneous fermentation process to produce flour with optimal characteristics as a raw material rice analog formulations.

## II. MATERIAL AND METHODS

*A. Materials*

Nagara bean take from Hulu Sungai Selatan districts exactly Nagara area, South Kalimantan Indonesia

*B. Methods of Spontaneous Fermentation*

Nagara beans soaked with water at a ratio of 1: 4 (w / v). Spontaneous fermentation with some variation wet fermentation periods (24 hours, 48 hours, 72 hours, 96 hours and 120 hours), by changing the soaking water everyday and form a fermented material that are whole bean (with hull), and grits. The fermented beans are cleaned from the hull, dried at a temperature of 60°C for 48 hours, then powdered and sieved to 80 mesh size. Nagara beans was tested amino acid composition using HPLC. Flour obtained by testing the moisture content (oven method) protein content (method Kjeldhal) bulk density, water absorption capacity, swelling volume and solubility.

*C. Bulk Density*

Bulk density was measured using a measuring cup. Samples to be measured, weighed as much as 10 g. Then put in a 50 mL measuring cup and readable volume. Bulk density is calculated as the ratio of the sample weight by volume of the sample read on the measuring cup.

$$\text{Bulk density } (g/cm^3) = \frac{\text{weight sample (g)}}{\text{volume}(cm^3)}$$

*D. Water Absorption Capacity*

Centrifuge tube filled with 2 g of flour samples were weighed and the weight of the sample tube (a), then added 9 mL of distilled water and vortex. Furthermore allowed to stand for 30 minutes and then centrifuged at 3000 rpm for 15 minutes, decanted and weighed (b).

Water absorption capacity (% db)

$$\text{Water absorption capacity (\% db)} = \frac{b-a}{ms\ (1-mc)} \times 100\%$$

a = weight of sample + weight sentrifuge tube (g)
b = weight of wet sample + weight sentrifuge tube (g)
ms = weight sample
mc = moisture content

*E. Swelling Volume (g/g db) and Solubility (% db)*

Swelling volume is determined by weighing as much as 0.35 g of flour are then added water to 12.5 mL centrifuge tube. Furthermore the solution have been vortex and then heated in a water bath with a temperature of 92.5°C and vortex once every 5 minutes for 10 minutes. Furthermore, the solution was cooled in ice water for 1 minute and at 25°C for 15 minutes. Then the solution was centrifuged at a speed of 3600 rpm for 15 minutes. Gel that is formed is measured in volume and is expressed as a swelling volume in unit g/g (db). Solubility is obtained by pouring the resulting supernatant into a cup that has been known weighed and dried at 110°C for overnight. Solubility is calculated by the following formula :

$$\text{Solubility (\%db)} = \frac{w1}{wdm} \times 100\%$$

$$\text{Water swelling volume (g/g db)} = \frac{w2}{ms\ (1-mc)}$$

w1= weight of supernatant (g)
w2 = weight of formed gel (g)
wdm= ms(1-mc)
ms = weight sample
mc = moisture content

*F. Statistical Analysis*

The data are analyze by using analysis of variance (ANOVA) and if there is real effect then continued with Duncan test (DMRT) at a rate of error of 5%.

## III RESULT AND DISCUSSION

*A. Composition Amino Acid in Nagara Bean*

Nagara bean is one type of bean with high carbohydrate and protein. Quality protein in a material can be seen from the composition of amino acids in it. The results showed that Nagara bean contain amino acids glutamate (2,183%), amino acid composition are presented in Table I.

TABLE I
AMINO ACID COMPOSITION IN NAGARA BEAN

| Amino Acid | Total (%) |
|---|---|
| Aspartic acid | 0.913 |
| Glutamic acid | 2.182 |
| Serine | 0.578 |
| Glycine | 0.258 |
| Histidine | 0.826 |
| Arginine | 0.584 |
| Threonine | 0.282 |
| Alanine | 0.116 |
| Proline | 0.196 |

| Amino Acid | Total (%) |
|---|---|
| Tyrosine | 0.218 |
| Valine | 0.734 |
| Methionine | 0.791 |
| Cysteine | 0.321 |
| Isoleucine | 0.393 |
| Leucine | 0.775 |
| Phenilalanine | 0.417 |
| Lysine | 0.438 |
| Tryptofan | - |

The **c**haracteristics of Nagara beans flour can be better obtained either by way of a natural modification through a process of spontaneous fermentation. Spontaneous fermentation is fermentation which does not add inoculum or starter from the outside, and the process is very influenced by the length of fermentation which is done. During fermentation, it is expected to occur fragmentation of starch granules that will affect the characteristics of the resulting flour. During fermentation, lactic acid bacteria grow and produce organic acids especially lactic acid. Subagio (2006) stated that during the fermentation of starch, there are changes in characteristics such as solubility, rehydration value, aroma and color. In this study, the fermentation is done spontaneously by soaking Nagara bean in the form of whole beans and grits with a long fermentation time 24 hours to 120 hours. The test results are presented in Table II.

TABLE II
THE CHARACTERISTICS OF SPONTANEOUS FERMENTED NAGARA BEAN FLOUR

| Treatments | | Parameter | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Size Bean | Fermentation Periods (hours) | Yields (%) | Moisture Content (%) | Protein (%) | Bulk Density (g/cm$^3$) | Water Absorption Capacity (% db) | Swelling Volume (% g/g db) | Solubility (% db) |
| Whole | 24 | 57.0±7.0 | 7.9±0.2 | 22.6±0.9$^{ab}$ | 0.46±0.0 | 142.2±8.2$^{a}$ | 553.6±11.2 | 24.4±2.0$^{d}$ |
| | 48 | 58.0±14.1 | 6.8±0.9 | 22.7±1.9$^{ab}$ | 0.50±0.0 | 177.4±14.8$^{b}$ | 522.1±69.9 | 16.9±7.7$^{d}$ |
| | 72 | 57.5±3.5 | 6.8±1.7 | 24.2±0.6$^{b}$ | 0.49±0.0 | 200.5±11.6$^{cd}$ | 518.1±5.6 | 10.6±1.3$^{cd}$ |
| | 96 | 45.5±7.7 | 5.9±0.6 | 23.1±1.0$^{ab}$ | 0.50±0.0 | 204.6±6.2$^{cd}$ | 530.8±72.3 | 7.1±0.0$^{ab}$ |
| | 120 | 40.0±2.8 | 5.2±1.1 | 22.6±0.2$^{ab}$ | 0.51±0.0 | 193.4±5.1$^{bcd}$ | 711.1±33.8 | 5.9±0.6$^{ab}$ |
| Grits | 24 | 60.5±4.9 | 6.3±0.2 | 24.3±2.5$^{b}$ | 0.50±0.0 | 186.5±5.9$^{bc}$ | 552.5±80.4 | 12.0±2.2$^{c}$ |
| | 48 | 59.5±12.0 | 6.3±0.3 | 23.9±0.7$^{b}$ | 0.52±0.0 | 201.1±3.1$^{cd}$ | 554.7±116.0 | 8.3±3.3$^{bc}$ |
| | 72 | 53.5±2.1 | 5.9±1.9 | 21.0±0.9$^{a}$ | 0.51±0.0 | 202.1±3.3$^{cd}$ | 541.9±38.3 | 6.5±0.9$^{ab}$ |
| | 96 | 51.5±0.7 | 5.8±0.5 | 22.2±0.8$^{ab}$ | 0.49±0.0 | 208.8±2.7$^{d}$ | 554.1±16.6 | 6.5±0.3$^{ab}$ |
| | 120 | 51.5±0.7 | 5.3±0.0 | 21.5±0.2$^{a}$ | 0.50±0.0 | 190.3±8.7$^{bcd}$ | 693.5±49.7 | 5.2±0.0$^{a}$ |

*numbers followed by the same letter are not significantly different ($\alpha$ 5%)

*B. Yields*

In a process, determining the yields are very important as a measure of the performance of processes. The result of variance analysis ($\alpha$ 5%) showed that the size of bean and periods of fermentation had no significant effect on yield of Nagara bean flour. In the process of spontaneous fermentation Nagara bean, the average yield is 53.45% which produced flour yield of grits size is relatively higher than the size of the whole, and the longer of the fermentation flour yield will decrease. It was caused by the longer of contact time, the process of absorption of water into the matrix of Nagara bean is increased, so did the performance fragmentation of lactic acid from lactic acid bacteria in degrading cellulose structure is intensified. So it makes the texture of Nagara bean increasingly softened, and the washing process will lead to losses. The process of softening Nagara beans is larger after fermentation periods 72 hours. In whole bean sizes, after the fermentation process, the hull was stripped, after 72 hours of fermentation the beans have tender texture, very soft, so when there were washed, the beans will be destroyed and lot of starch dissolved in water. While in the grits size, the hull already partially separated from the beans, the emphasis on beans to remove the skin is relatively small, and has lower structural damage, so the loses of it is smaller.

*C. Moisture Content*

The determination of moisture content was aimed to determine the water content in the fermented nagara bean flour. Nagara bean flour moisture content ranging from 4.42 to 8.04% with an average value of 6.20%. The result of variance analysis ($\alpha$ 5%) showed that the size of bean and periods of fermentation had no significant effect on moisture content produced nagara bean flour.

On the whole size beans, flour moisture content ranges from 5-7.5% while the grits size ranges from 5-6% water content. The size of bean which are still intact beans are covered by the hull so that the rate of diffusion of water into the bean will be slower than bean in the form of grits. Size grits expand the surface area so contact between the beans with water and organic acids will degrade performance granular structure easier, so that the water content in the drying process easily evaporated. If fermentation process so longer, so the process of fragmentation of granules is getting bigger, so during the drying process, the water content of flour will decrease.

*D. Crude Protein Content*

The crude protein content of fermented Nagara bean flour is high enough range 21.02 to 24.20%. Bean in general are a source of protein, with the fermentation process through soaking, it will be possible for protein hydrolysis become simpler components namely peptides so that its availability will be higher. The lactic acid produced from lactic bacteria that grow spontaneously will accelerate the process of hydrolysis of proteins.

The result of variance analysis ($\alpha$ 5%) showed the interaction size bean and periods of fermentation significantly affected protein content of modified Nagara bean flour. The highest protein content generated on the grits were fermented for 24 hours (24.29%) and did not differ with whole bean with a fermentation periods 72 hours (24.20%). The size of grits is suspected hydrolysis or breakdown protein by proteolytic enzymes and acids that occurs faster than the size of the whole. On grits size, the pH of fermentation media decrease faster, on 24 hours fermentation it reaches pH 5, while on the whole size, on 24 hours fermentation it reaches pH 6-7.

Supposedly on grits size allows lactic bacteria grow rapidly due to the availability of nutrients from the beans and more easily obtained compare with whole size bean.

### *E. Bulk Density*

Bulk density is needed to determine the weight of material per unit volume. Modified Nagara bean flour had a softer texture than unspontaneous modification. Bulk density of modified Nagara bean flour ranges from 0.46 to 0.52 $g/cm^3$. The result of variance analysis ($\alpha$ 5%) showed Nagara bean size, fermentation periods and interaction of both have not significant effect on the bulk density of the Nagara bean flour. Smaller bulk density of material will facilitate the packaging process and transport.

### *F. Water Absorption Capasity*

Starch granules which are the main components can be inflated by soaking in cold water. In cold water, the water absorption capacity is limited, but when starch is heated, the thermal energy will break the hydrogen bonds so the surface area for absorption of water become larger and starch granules will absorb more water.

The result of variance analysis ($\alpha$ 5%) showed that the interaction size nagara bean and fermentation periods significantly affect the water absorption capacity of the Nagara bean flour produced. The highest rate of water absorption is obtained in the 96 hours fermentation of grits size and relative no different with fermentation periods 48 and 72 hours. Supposedly greater water absorption is due to the fragmentation of the starch and protein during spontaneous fermentation process so that the binding of water becomes greater. According Etudaiye *et al.* (2009), during fermentation, the proteolytic activity of the microorganisms cause additional polar groups on the starch granules, and the addition of polar groups will increase flour hidrofility.

Differences in the absorption of water is caused due to differences in surface area, that is broken bean and protein content (Sabularse *et al*. 1991). Water absorption capacity tends to increase in fermentation time up to 96 hours, while at fermentation periods 120 hours tend to lower water absorption. Grits size at 24 hours fermentation had grater water absorption capacity than the size of the whole, it is presumably because the larger surface area. Fragmentation of starches and proteins occur more quickly so that the absorption of water more quickly too.

### *G. Swelling Volume*

Swelling volume indicates the level of development of starch granules when interacting with water, the more water is absorbed, the rate of granule development will be even greater. The result of variance analysis ($\alpha$ 5%) showed that the fermentation periods significantly affect swelling volume of the fermented bean flour. Swelling volume is likely to increase up to 120 hours fermentation periods. Leach *et al.* (1959) stated swelling volume is influenced by the strength of the bond between the granular network. Lactic acid bacteria produce amylase will attack amorphous areas of starch granules, the performance is affected by hydrolytic enzymes and organic acids produced during fermentation. During heating in water, the starch granules occur gelatinization and will expand and partially dissolved causing a viscous solution. Rigid structure that would be difficult to expand because the matrix is still strong so the water absorption becomes limited.

### *H. Solubility*

The solubility of the modified Nagara bean flour ranges from 5.15 – 24.38 %db. The result of variance analysis ($\alpha$ 5%) showed that the interaction of nagara bean and fermentation periods have significant effect on the solubility of flour. On the size of grits solubility in water flour is relatively lower than full size and the longer of the fermentation, the smaller of the solubility flour. Swelling of the starch granules above the gelatinization temperature is accompanied by washing polisaccharides soluble. Amylose which is polar, if much amylose component out of the granules, the solubility increases. With the longer the fermentation, suspected amylose exposure is reduced so that the solubility decreases. The amount of dissolved amylose is a function of the organization's internal granular starch (Walter *et al*. 2000).

## IV CONCLUSION

Treatment of grits size produce flour yield, protein content, water absorption capacity, and the swelling volume greater than others. Fermentation time up to 120 hours tend to lower flour yield and protein content, while the water absorption capacity tend to increase up to 96 hours fermentation and swelling volumer tends to be stable up to 96 hours fermentation periods.

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