

## Physicochemical properties of *Bruguiera gymnorrhiza* flour (BGF)

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### Abstract

This research aims to study the physicochemical properties of *Bruguiera gymnorrhiza* flour (BGF). The peeled BG fruit was immersed in ash suspension in water (10% w/v) for 24 hours, then sundried, milled, and sieved through membrane sized 100 mesh. Particle size of BGF produced as described are of 150  $\mu\text{m}$ , brown in colour, of low viscosity and adhesiveness (CR:30.25), low amylose content (14.23%), had low protein (4.82%) and fat (0.24%) content, and high crude fiber content (7.46%). Tanin content of BG fruit was successfully reduced by pretreatment in this study (19700 ppm to 8500 ppm). HCN content in BG fruit were under the detection limits of our method to be determined. Results of the present study suggests futher studies on the use of BGF for developing flat bread ingredients.

### Keywords

*Bruguiera gymnorrhiza*  
Flour  
Physicochemical properties

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### Introduction

Mangrove is a group of plants living in a tidal swamp ecosystem, which protects the land from the erosion of sea waves, with great resistance to environments with high humidity and salinity. There are cultivated species, such as *Avicennia* sp., *Rhizophora* sp., *Bruguiera* sp. and *Sonneratia* sp. *Bruguiera gymnorrhiza* (BG), locally known as Lindur Fruit in Indonesia, are also cultivated in tropical areas such as South Africa, East Africa, Madagascar, South Asia, Southeast Asia and Australia. Fruit production begins in two-year-old specimens with 15 cm to 25 cm in length and 2 cm in diameter.

The whole *Bruguiera gymnorrhiza* fruit consists of about  $80 \pm 1.75\%$  of edible fruit and  $20 \pm 1.75\%$  of peel. So far, BG is processed into *Bruguiera gymnorrhiza* flour (BGF), where pretreatment is required to reduce the content of antinutritional substances, such as HCN and tannin. HCN is a result of enzymatic reaction after maceration (Butler, 1965). Tannins are polyphenolic compounds found in most plants and are generally thought to function as chemical defenses against pathogens and herbivory (Gedir *et al.*, 2005); tannins have antioxidant and antibacterial activities (Sung *et al.* 2012).

Excess HCN consumption can be poisonous because HCN inhibits cytochrome-oxidase in human aerobic respiration and inhibition of this enzyme

may cause asthma (Cooper and Brown, 2008). Other conditions caused by excessive HCN are headache, dizzy and diarrhea (Nhassico *et al.*, 2008). Tannin is actually required for free-radical scavenging in human body, but it can be a carcinogen as well as intensify fruit's bitterness in excessive concentration. HCN and tannin are volatile water soluble substances (Cumbana *et al.*, 2007), therefore a previous study suggested using water rinsing and/or blanching to reduce the content of HCN and tannin (Subandriyo and Setianingsih, 2016). However, the water rinsing method seems inefficient because it required repetition.

In order to reduce the use of water in pretreating BG, a more effective method is needed. The prior study reveals that the use of ash suspension is able to reduce the amount of HCN and tannin by immersing the sample containing HCN and tannin in the ash suspension, because ash contains  $\text{SiO}_2$ ,  $\text{Al}_2\text{O}_3$ ,  $\text{Fe}_2\text{O}_3$ ,  $\text{CaO}$ ,  $\text{K}_2\text{O}$ , carbon and unburnt residue which can absorb HCN and tannin (Ferrer-Mairal *et al.*, 2012).

In spite of BGF consume by population in coastal areas and evidence from researches, the use of this flour needs to be optimized because current studies on the physicochemical properties and suitability for consume of this flour are incomplete (Sulistyowati, 2012; Subandriyo and Setianingsih, 2016). Therefore, the present study was performed to determine the physicochemical properties of BGF.

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## Materials and Methods

### Materials

BG fruit with brown colour was harvested in Mangrove Conservation (Wonorejo, Surabaya East Java Indonesia). The fruit was then carried to the processing laboratory and directly processed, wood ash was purchased from the local market (Pucang, Surabaya East Java Indonesia), Sieve with mesh no. 100 (membrane size of 150  $\mu\text{m}$  cut-off) and coffee grinder.

### Preparation of BGF

BGF was produced according to Sulistyowati and Kumalaningsih (2012) with slight modification. BG fruit was blanched at 80°C - 90°C for 5 minutes, and then the fruit was peeled and sliced by using an aluminium knife and then soaked in 10% (w/v) ash-water suspension until the fruit was fully soaked for 24 hr. The fruit was sun dried until fully dry. The dried BG fruit was then milled and sieved through 100 mesh filter medium.

### Proximate analysis

Proximate composition of protein, fat, moisture content, ash content, amylose and HCN were analyzed using reference method of AOAC (2004), and carbohydrate was analyzed using by difference method. All proximate components were analyzed in triplicate.

### Determination of flour density, Carr index, Hausner ratio

Bulk density was measured by method Okezie and Bello (1988). BGF was loaded into a beaker glass for volume and weight determination and bulk density was calculated. Tapped density was carried out by tapping several times until persistent volume was obtained, and weighing the flour at that moment. Density was expressed as BGF mass (g) per volume (mL). Hausner ratio and Carr index were analyzed using this following equation:

$$\text{Hausner ratio (HI)} = \frac{\text{Tapped density (g/ml)}}{\text{Bulk density (g/ml)}}$$

$$\text{Carr Index (CI)} = \frac{\text{Tapped density (g/ml)} - \text{Bulk density (g/ml)}}{\text{Tapped density (g/ml)}} \times 100$$

### Gel strength

Gel strength was analyzed using methods described by Hsu *et al.* (2003). BGF suspension 7.00% w/v was made by dispersing BGF into water followed by homogenization with a magnetic stirrer. The suspension was then heated in a tube

at 90°C for 20 minutes and cooled in a refrigerator for 12 – 24 hr. The gel was then analyzed using a texture analyzer (Stevens-LFRA Texture Analyser®, Mechtric-Stevens), with cylindric probe of 0.1923 inch diameter, at velocity of 2.00 mm/sec and 15mm penetration.

### Gel hydration properties

Water absorption index (WAI), water solubility index (WSI) and swelling power (SP) were analyzed according to Toyokawa *et al.* (1989), with slight modification. 100 mg BGF ( $W_i$ ) was dispersed in 2.0 mL water, and then the dispersion was heated at the temperature of 60°C, 75°C and 90°C for 10 minutes in a thermostat water bath. Furthermore, BGF suspension was cooled until 4°C and centrifuged at 3,500 rpm for 15 minutes. Supernatant was recovered, and dried at 105°C until constant weight ( $W_s$ ); residue was also weighed ( $W_r$ ). Analysis was carried in triplicate and results calculated according to the following equations:

$$\text{WAI} \left( \frac{\text{g}}{\text{g}} \right) = \frac{W_r}{W_i}$$

$$\text{WSI} \left( \frac{\text{g}}{\text{g}} \right) = \frac{W_s \times 100}{W_i}$$

$$\text{SP} \left( \frac{\text{g}}{\text{g}} \right) = \frac{W_r}{W_i - W_s}$$

### Pasting properties

Pasting properties of BGF was analyzed using a Rapid Visco Analyser (RVA-4, Newport Scientific Pty Ltd., Warriewood, NSW2102, Australia) with seven parameters including breakdown viscosity, final viscosity, setback viscosity, peak time, pasting temperature, trough viscosity and peak viscosity. In the aluminium can, 2.5 g of BGF was dispersed in 25 mL water. The mixture was then spinned (160 rpm) at 50°C for 1 mins, and heated to reach 95°C within 7.5 mins, and held at 95°C for 5 mins, and then cooled back to 50°C within 7.5 mins and held at 50°C for additional 2 mins (Farasara *et al.*, 2014).

### Analysis of colour

BGF color was determined by using a chromameter (Minolta Type CR-300, Japan) and measured  $L^*$ ,  $a^*$ , and  $b^*$ . The  $L^*$  scale ranges from 0 for black to 100 for white; the  $a^*$  scale extends from a negative value (green hue) to a positive value (red hue); and the  $b^*$  scale ranges from negative blue to positive yellow (Tharise *et al.*, 2014).

### BGF microstructure

The microstructure of the BGF and the profile of atoms composing BGF were investigated using



Figure 1. BGF (*Bruguiera gymnorrhiza* flour)

a Scanning Electron Microscopy with Energy-dispersive X-ray spectroscopy (ZEISS EVO® MA 10, Quantax Energy Dispersive X-ray).

## Results and Discussion

BGF has small particle size and is cohesive, resulting in a dense texture. Cohesiveness causes the flour to quickly collapse by tapping Abdullah and Geldart (1999). Hausner ratio and Carr index values reflected that BGF was fairly flowable and very cohesive. Hausner ratio and Carr index of powdered samples are also correlated with their density.

Brown colour of BGF was indicated by the values  $a^*$  and  $b^*$  in colour analysis (Figure 1, Table 1). The amount of tannin in BGF contributes to the brown colour (Kennedy *et al.*, 2006), because tannin is easily oxidized (Sims and Morris, 1986). The oxidation of tannin may occur at pretreatment in ash suspension or sun drying stages during flour making process. Sims and Morris (1986) stated that tannin can sharpen product colour when it is incorporated as ingredient. Therefore, BGF used as food ingredient may add brown colour to the final product.

According to results from pasting properties studies determined with rapid visco analyzer, BGF could not thicken a water solution containing because it had low final viscosity, breakdown viscosity and setback viscosity. The low breakdown viscosity of BGF suggests that BGF paste would have high stability, resistance to retrogradation and low water holding capacity. This finding was in line with prior study carried out by Shafie *et al.* (2016), where the use of flour with low breakdown viscosity results in more stable paste product.

BGF produced had low WSI because of its low protein content. WSI represents the amount of water-soluble components exposed in water (Filli *et al.*, 2010). Gel hydration properties of BGF increased with temperature, this finding was in line with

Table 1. Physical properties of BGF

Parameter	BGF
Yield (%w/w)	1.68 ± 0.76
Tapped density (g/mL)	0.76 ± 0.06
Bulk density (g/mL)	0.53 ± 0.02
Hausner ratio	1.44 ± 0.08
Carr Index	30.25 ± 3.88
Particle size (µm)	≤ 150.00 ± 0.00
Colour	
L <sup>a</sup>	51.77 ± 0.21
a <sup>a</sup>	17.00 ± 0.10
b <sup>a</sup>	17.40 ± 0.10
Gel strength (Newton)	0.10 ± 0.00
Pasting properties	
Breakdown Viscosity (cP)	112.50 ± 2.12
Final Viscosity (cP)	1243.50 ± 20.51
Setback Viscosity (cP)	329.00 ± 2.83
Peak time (min)	9.44 ± 0.23
Pasting Temperature (°C)	84.93 ± 0.04
Trough Viscosity (cP)	914.50 ± 17.68
Peak Viscosity (cP)	1027.00 ± 19.80

\*Data was expressed as means (n : 3) ± SD

NA : not analyzed

previous studies carried out by Bhat *et al.* (2016).

Amylose content may affect flour viscosity (Blazek and Copeland, 2008) and gel strength. Compared to wheat flour (Bhat *et al.*, 2016), BGF had lower amylose content and lower viscosity. Concordantly, the gel strength of BGF at 7% (w/v) was also very low. In bread making, pasting properties affect bread texture, and flour with high viscosity tends to produce bread with finer texture (Wani *et al.*, 2015). Gel strength is also a common indicator of food quality in hydrocolloids such as carrageenan, gelatin, amylose, and alginate.

The microstructure analysis (Figure 2) of BGF in the present study demonstrated that starch granules of BGF were of 150 µm cut-off because siever of 100 mesh was used. Starch granules sized 12 µm cut off are considered small (Sahlström *et al.*, 1998). The amount of small granules affects the water retention during baking since they bind water (Soulaka and Morrison, 1985; Sahlstrom *et al.*, 1998). Water tends to be associated with starch rather than with protein in flour where this phenomenon increases dough stiffness and reduces flow of the dough during fermentation and baking (Sahlström *et al.*, 1998).

According to EDX spectrum (Figure 3), BGF was rich in carbon and oxygen, but poor in nitrogen. The amount of carbon, oxygen, and nitrogen were related to the content of carbohydrate and protein in BGF. The higher carbon and oxygen atom interpreted

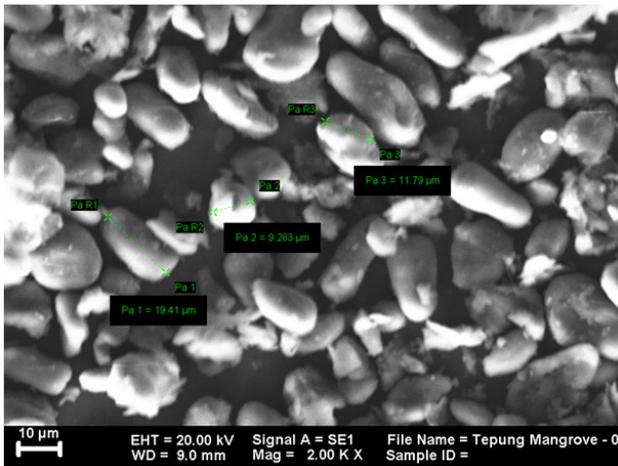


Figure 2. Microstructure properties of BGF, Pa 1, Pa 2 and Pa 3 represented starch granule's length, width and diameter respectively

through EDX spectrum represented the higher carbohydrate and low protein contained in BGF.

There is a challenge in utilizing BG fruit into flour, the yield gained was too low (1.68% w/w). The used method in this present study resulted by product as flour with higher particle than desired size ( $\leq 150.00 \mu\text{m}$ ). Consequently, the used method in this present study might require more intensive milling to enhance the yield. On the other hand, this finding recommended further study on the utilization of this flour in small pieces to get fine product with desired functionalities.

Chemical composition of BGF suggest BGF might have pro-health functionalities, such as antidiabetic and antioxidant properties because it contains crude fiber and tannins (Onyeneho and Hettiarachchy, 1992). Fiber is non digestible carbohydrate on the upper gastrointestinal track that will not contribute to the increasing of blood glucose while consumed. Tannins are polyphenol compounds able to form complexes with protein such as digestive enzymes and cause their inhibition (Ekop *et al.*, 2008). Therefore, tannins could be incorporated in food product for desired functional food reducing the risk of diabetes. Excess tannin, however, will result in adverse effects (Ekop *et al.*, 2008). Subandriyo and Setyaningsih (2016) stated that the acceptable daily intake of tannin is 560 mg/Kg body weight/d.

During boiling and immersion in ash suspension, the water colour changed from clear to opaque. Ferrer-Mairal *et al.* (2012) states that ash containing  $\text{SiO}_2$ ,  $\text{Al}_2\text{O}_3$ ,  $\text{Fe}_2\text{O}_3$ ,  $\text{CaO}$ ,  $\text{K}_2\text{O}$ , carbon and un-burnt residue of which can absorb tannins and HCN. Pretreatment method with ash applied in the present study successfully reduced tannin content and might have reduced HCN content as well. Fortuna *et al.* (2017) demonstrated that HCN reacted to CaCN

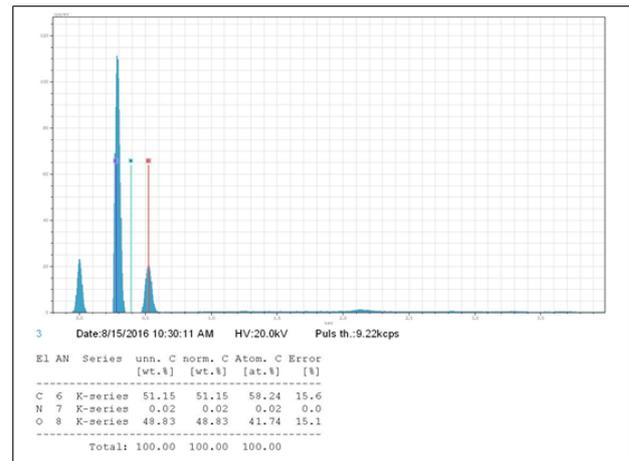


Figure 3. EDX spectrum of BGF demonstrating atoms composing BGF

and diffused to the used water during boiling and immersion. The use of high temperature for blanching might inactivate HCN linamarinase catalyzing HCN formation from cyanide glycoside after harvesting and evaporate HCN (Butler, 1965). The content of HCN in plant depends on the habitat where the plant grows and the maturity stage (Hue *et al.*, 2012). HCN levels were under detection limit in raw fruit and flour in our study.

The content of macronutrient is an important parameter for the evaluation of BGF as food. BGF could be categorised as low protein flour compared to wheat flour (Bhat *et al.*, 2016). A study on Taftoon bread (flat bread) carried out by Salehifar *et al.* (2010) reveals that flour with high protein reduces firming and staling of bread during storage, and the dough produced can not be able to sheet and expand under high temperature. Hence, the study on the use of BGF for flat bread making is needed because BGF can be categorized as flour with low protein content.

Compared to wheat flour produced by Bhat *et al.* (2016) where the fiber is 5.60%, BGF has high crude fiber (7.46%). Fiber plays an important role in diet management for diabetes (Onyeneho and Hettiarachchy, 1992). Some industries have developed high fiber content food product to contribute diabetes diet management. In bread making, fiber can increase final product hardness but reduce bread volume, because fiber restricts the expansion of gas cells. Fiber also causes appearance of dark colour on crust and crumb of the bread (Amir *et al.*, 2012). These phenomena are desired by many food industries producing flat bread, cookies and biscuits.

BGF contained higher ash content than wheat flour, which suggests higher mineral content in BGF than in wheat flour. Minerals are needed to sustain living organisms mainly as cofactors for enzyme activity.

## Conclusion

BGF produced in this present study had low protein, fat and amylose content, and high crude fiber with brown colour. This flour contained tannin amounts within acceptable daily intake. HCN levels in fruit and flour were under detection limit. This study suggests further studies on the use of BGF in developing flat bread ingredients based on its physicochemical properties.

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