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by Eko Nurcahya Dewi , Retno Ayu Kurniasih, Lukita Purnamayati

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Nutritional and Antinutritional Properties of *lindur* (*Bruguiera gymnorrhiza*) Fruits Flour from Different Pre-Treatments

Eko Nurcahya Dewi*, Retno Ayu Kurniasih, and Lukita Purnamayati

Department of Fisheries Technology, Faculty of Fisheries and Marine Science, Diponegoro University, Semarang, 50275, Indonesia

Background: *lindur* (*Bruguiera* sp) fruits are rich in nutrition and bioactive compound, but also had several anti-nutritional factors like tannins, saponins, and hydrogen cyanide (HCN). The aim of this study was to investigate the effect of pre-treatments on the preparation of *lindur* fruits flour to the nutritional and antinutritional properties, and to investigate the best pre-treatment to decrease the antinutritional properties in *lindur* fruit flour. *Method*: The pre-treatments were conducted by soaking the fruit in water, saturated salt solution, $CaCO_3$ solution, and rice husk ash solution. Nutrition of *lindur* flour was determined in terms of proximate composition and crude fiber. Antinutritional factors of the *lindur* flour were determined of tannin, saponin, and hydrogen cyanide (HCN). *Results*: Pre-treatment decreased the moisture content but increased protein, lipid, carbohydrate, and crude fiber of *lindur* flour. By soaking into saturated salt, nutrition could produce lowest moisture yet highest protein, carbohydrate, and crude fiber content. Pre-treatment with soaking into saturated salt solution also showed the higher reduction of tannin and HCN content compared to soaking into water, $CaCO_3$ solution, and rice husk ash solution. The *lindur* fruits flour soaking into saturated salt solution has a tannin content (0.766 ± 0.070)% and HCN content (231.705 ± 6.004) ppm. Meanwhile, pre-treatment by soaking in water, saturated salt solution, and $CaCO_3$ solution did not affect the saponin content of *lindur* flour. *Conclusion*: Different soaking method affects the nutritional value and antinutritional properties of *lindur* flour. The soaking treatment into saturated salt solution was the best pre-treatment method.

Keywords: Antinutritional, *lindur*, Nutritional, Pre-Treatments.

1. INTRODUCTION

Mangrove forests are widely spread in the coastal areas of Indonesia, nearly 25% of the world's mangrove forests. *lindur* (*Burguiera gymnorrhiza*) is one of the mangrove species found in Indonesia.[1] *lindur* fruits have long beans that can be used for human consumption and as a traditional medicine. The high carbohydrate content of the *lindur* fruits make the fruits become a new food source.[2–5] Although *lindur* fruits are rich in bioactive compound, but their use in food is still limited because of the presence of several antinutritional factors like tannins, saponins, and hydrogen cyanide (HCN).

Tannins are phenolic compounds that can interfere with iron absorption in the gastro-intestinal tract, thus decreasing the bioavailability of iron. Tannins are responsible for the taste of food and drinks.[6–8] Saponin has a bitter taste to form a stable foam in water, and cannot be removed with the addition of acid. Some saponin is toxic and known as sapotoxin.[9–11] While HCN is responsible for tissue hypoxia. Exposure of HCN causes respiratory cardiovascular, neurological, and thyroid defects.[12] These toxic factors present in *lindur* fruits must be removed or reduced in order to utilize its entire nutritional potential.

The methods for reduction of antinutritional factors of a food source can be done through a heating process. Antinutritional factors of the food source are unstable to heat, so it can be reduced with heat treatments and the use of chemicals, such as hydrochloric acid, acetic acid, and calcium hydroxide.[13]

The community of the northern coast of Jawa island, Indonesia, has been consuming this fruit for a long time. They soaked the fruit in water and rice husk ash solution before processing. Therefore, further study is needed to determine the different soaking solution toward the nutritional and antinutritional value of *lindur* flour. The aim of this study was to investigate the effect of pre-treatments on preparation *lindur* fruit flour to the nutritional and antinutritional properties and also to investigate the best pre-treatment to reduce the antinutritional properties of *lindur* fruit flour. In this study, the *lindur* fruits were soaked in water, saturated salt solution, $CaCO_3$ solution, and rice husk

*Author to whom correspondence should be addressed.

ash solution for pre-treatments on preparation *lindur* fruits flour. The proximate composition, crude fiber, tannin content, saponin content, and HCN content of *lindur* fruits flour were analyzed.

2. METHOD

2.1. Material

The *lindur* (*Bruguiera gymnorrhiza*) fruits from the north coast of Kendal, Central Java, Indonesia, were used as a primary raw material. Water, saturated salt solution, $CaCO_3$ solution, and rice husk ash solution were used as reducing agent of antinutritional factors.

2.2. *Lindur* Fruits Flour Preparation

The *lindur* fruits were peeled and boiled at 100 °C in an aqueous solution for an hour. Each of samples was treated randomly to four different treatments were soaked on water (A_1), saturated salt solution (A_2), $CaCO_3$ solution (A_3), and rice husk ash solution (A_4). The fresh *lindur* fruits were used as control (A_0). Each of solutions made with the ratio of material (salt, $CaCO_3$, and rice husk ash) and distilled water of 1:10 (w/v). The fruits were soaked in the four different solutions for 72 hours with a ratio 1:3 (w/v). After finished soaking, the fruits are sliced, dried, and then were ground with a miller 80 in mesh size.

2.3. Proximate Composition and Crude Fiber

The proximate composition (moisture, protein, lipid, ash, and carbohydrate content) and crude fiber of *lindur* fruits flour were carried out in triplicate using the methods described by AOAC.[14]

2.4. Tannin Content

Lindur fruit flour (1,5 g) was added 50 mL of aquadest at 60 °C and set aside for 30 minutes. The sample solution was filtered and aquadest was added until 250 ml. The sample (25 mL) was added 20 mL of indigocarmin solution and titrated using $KmnO_4$ 0,1 N solution until the royal blue fades to a light green. Then drop-wise titration until the lime green changes to yellow. The titrant volume needed was noted as A mL.

The blank titration using 25 mL aquades was added 20 mL of indigocarmin solution. This titrant volume noted as B mL. The tannin content calculated using formula.[1,6]

$$\text{Tanin content}(\%) = \frac{(10(A-B)\times N\times 0.00416)}{\text{Sample(g)}}\times 100\%$$

2.5. Saponin Content

The *lindur* fruits flour (20 g) were put into a conical flask and 100 ml of 20% ethanol was added. The samples were heated with a hot water bath for 4 h with continuous stirring at 55 °C. The solution was filtered and the residue was extracted again with another 200 ml of 20% ethanol. The extracts were reduced to 40 ml at about 90 °C. The concentrate was put into a 250 ml separatory funnel and 20 ml of diethyl ether were added and shaken. The solution layer was recovered while the ether layer was discarded then 60 ml of *n*-butanol was added. The mixture was washed twice with 10 ml of 5% aqueous sodium chloride. The solution was evaporated in a water bath. After that, the samples were dried in the oven to a constant weight. The saponin content was calculated as mg/100 g.[8]

2.6. HCN Content

HCN content determination was done with the iso-nicotinic acid-3-methyl-1-phenyl-5-pyrazolone. The HCN was converted to cyanogen chloride with an aqueous solution of chloramine-*T*, and then absorbed by sodium hydroxide. The reaction of the cyanogen chloride with pyridine and then with a 3-methyl-1-phenyl-5-pyrazolone reagent results in a colored complex, which was analyzed by a spectrophotometer at 638 nm.[15]

2.7. Statistical Analysis

The mean values of multiple groups were analyzed by one-way analysis of variance (ANOVA). To determine the difference between treatments, Duncan test was conducted. P value of $<0,05$ was classified as statistically significant.

3. RESULTS

The proximate composition (%) and crude fiber (%) of *lindur* fruits flour were presented in Table I. The study results showed that the proximate composition of the fresh *lindur* fruits were moisture content (52.221 ± 0.710)%, protein content (4.433 ± 0.066)%, lipid content (0.896 ± 0.074)%, ash content (4.550 ± 0.224)%, carbohydrate content (82.266 ± 0.438)%, and crude fiber (7.870 ± 0.247)%. Pre-treatments produce *lindur* flour with moisture content between (10.969 ± 0.241)% to (12.767 ± 0.079)%, protein content (4.027 ± 0.411)% to (5.714 ± 0.452)%, lipid content (0.440 ± 0.062)% to (0.932 ± 0.099)%, ash content (0.944 ± 0.024)% to (4.364 ± 0.469)%, carbohydrate content (83.475 ± 0.058)% to (90.824 ± 0.836)%, and crude fiber content (8.861 ± 0.021)% to (14.770 ± 0.063)%.

Fresh *lindur* had tannin content of (1.221 ± 0.007)%, saponin content (0.030 ± 0.013)%, and HCN content (1498.907 ± 10.340) ppm. The antinutritional properties of the *lindur* fruits flour were presented in Table II. The results showed that the *lindur* fruits flour had a tannin content (0.766 ± 0.070 to 1.648 ±

Table I. Nutritional of the *lindur* fruits flour.

Sample	Moisture content (%)	Protein content (%db)	Lipid content (%db)	Ash content (%db)	Carbohydrate content (%db)	Crude fiber (%db)
A_0	52.221 ± 0.710^a	4.433 ± 0.066^a	0.896 ± 0.074^a	4.550 ± 0.224^a	82.266 ± 0.438^a	7.870 ± 0.247^a
A_1	12.767 ± 0.079^b	5.265 ± 0.026^b	0.440 ± 0.062^b	0.944 ± 0.024^b	83.475 ± 0.058^b	9.872 ± 0.042^b
A_2	10.969 ± 0.241^c	5.714 ± 0.452^b	0.932 ± 0.099^a	3.073 ± 0.125^c	90.393 ± 0.686^c	14.770 ± 0.063^c
A_3	12.225 ± 0.835^b	4.027 ± 0.411^a	0.845 ± 0.079^a	4.364 ± 0.469^a	90.824 ± 0.836^c	14.645 ± 0.123^c
A_4	11.266 ± 0.045^c	5.341 ± 0.048^b	0.857 ± 0.026^a	0.948 ± 0.033^b	83.995 ± 0.054^b	8.861 ± 0.021^d

Notes: Mean of three replications ± standard deviation. Means in the same column and different letters are significantly different ($P < 0.05$).

Table II. Antinutritional of the *lindur* fruits flour.

Sample	Tannin content (%)	Saponin content (%)	HCN content (ppm)
A_0	1.221 ± 0.007^{a}	0.030 ± 0.013^{a}	1498.907 ± 10.340^{a}
A_1	1.282 ± 0.001^{ac}	0.067 ± 0.009^{b}	202.773 ± 16.583^{b}
A_2	0.766 ± 0.070^{b}	0.065 ± 0.010^{b}	231.705 ± 6.004^{c}
A_3	1.358 ± 0.002^{c}	0.056 ± 0.011^{b}	489.304 ± 12.434^{d}
A_4	1.648 ± 0.002^{d}	0.102 ± 0.020^{c}	375.493 ± 10.757^{e}

Notes: Mean of three replications ± standard deviation. Means in the same column and different letters are significantly different ($P < 0.05$).

0.002)%, saponin content (0.056±0.011 to 0.102±0.020)%, and HCN content (202.773 ± 16.583 to 489.304 ± 12.434) ppm.

4. DISCUSSION

4.1. Proximate Composition and Crude Fiber

The moisture content of fresh *lindur* fruits in this study was different from the earlier reports which the moisture content of fresh *lindur* fruits was 65.18% and has been dried decreased to 9.18%.[5] There was the significant difference in the moisture content of *lindur* fruits flour as affected by the different soaking agent which soaking on saturated salt solution showed the *lindur* fruits flour had the lowest moisture content. The moisture content in *lindur* fruits flour was used for microbiological spoilage to grow.[16]

The results showed that protein content of *lindur* fruit flour was lower than protein content of wheat bran 9.6%, rice bran 11.8%, barley bran 11.9%, and oat bran 15.0% in the study Kaur et al.[6] Thus, *lindur* fruits flour is not a good source of protein. The high protein content showed that it might offer an affordable source of protein to counteract protein malnutrition.[17] The pre-treatment of soaking in different solutions can increase the protein content of *lindur* fruits flour. According to Nurjanah et al.,[5] there was an increase in protein content due to the reduced moisture content, thereby increasing the proportion of protein. The lipid content of *lindur* fruits flour was lower than another source of carbohydrate, such as wheat bran 4.1%, rice bran 19.3%, barley bran 4.2%, and oat bran 10.6%.[6]

Soaking treatment in water, saturated salt solution, and rice husk ash solution can reduce the ash content of *lindur* fruits content. This related to how much water can be drawn out of the *lindur* fruits while soaking. The water coming out of the *lindur* fruits carry minerals. Ash content reflects the quantity of mineral matter present in the flour.[15]

Based on research of Kaur et al.,[6] carbohydrate content in wheat bran 60.5%, rice bran 36.6%, barley bran 51.8%, and oat bran 55.6%. The high carbohydrate content shows that it can serve as a good source of energy.[18] Differences in the nutritional composition can be influenced by the environment or habitat and species.[19]

The highest crude fiber was caused by the soaking *lindur* fruits in a saturated salt solution and $CaCO_3$ solution. Crude fiber cannot be digested by human, it plays a useful role in providing roughage that aids digestion and reduces the accumulation of carcinogen in the body.[18] In the research by Kaur et al.[16] wheat bran, rice bran, barley bran, and oat bran has a crude fiber 7.75%, 11.5%, 14.9%, and 3.31%; respectively.

4.2. Tannin Content

When compared to other carbohydrate sources, *lindur* fruits flour produced higher tannin content, despite soaking. Kaur et al.[6] reported that tannin content in different defatted cereals are wheat bran 2.22 (mg/g) or 0.222%, rice bran 0.58 (mg/g) or 0.58%, barley bran 2.73 (mg/g) or 2.73%, and oat bran 5.17 (mg/g) or 0.517%. The tannins are concentrated in the seeds coat, so that the de-hulling can reduce tannin content of cereals bean.

The lowest tannin content in *lindur* fruits flour was caused by soaking in a saturated salt solution. The tannin content reduction can be influenced by the presence of the electrolyte, such as NaCl. It is caused due to the electrostatic force between the adsorbent surface and adsorbate ions, thereby causing adsorption of tannins.[20] The reduction of tannin also because these compounds are heat labile and degrade upon heat treatment. Soaking also showed a significant reduction in tannin content. This reduction could be due to leaching of tannin into the soaked solution.[15]

4.3. Saponin Content

Lindur fruits flour in this study contains saponins higher than saponin content of fresh *lindur* fruits. This is in contrast with the statement of Kaur et al.[13] in which the saponins can be lost due to heating because they are thermo-labile. Soaking can also lead leaching out of saponin into the water through diffusion.

The treatment of soaking in saturated salt solution and solution causes saponin content in *lindur* fruits flour lower than the treatment of soaking in a rice husk ash solution. This is presumably due to salt and $CaCO_3$ have the ability to adsorpt of the saponins. Li et al.[20] stated that the inorganic salts can influence the adsorption of organic compound, such as saponins.

4.4. HCN Content

The treatment of soaking in water, produced *lindur* fruits flour with lowest HCN content. The similar study result demonstrated by Soetan[21] which phytochemicals contain (including HCN) in *Lablab purpureus* can be reduced by soaking, heating, and boiling in water for an extended period of time. HCN is very volatile and very poisonous liquid, which can evaporate at 26 °C. When the temperature increases, the reduced nitrogen in HCN is equal to that in the formed N_2.[22]

The results showed that the content of HCN in *lindur* fruits flour was higher than HCN content in cassava dried (4.20±0.01) mg/100 g or (42±0.01) ppm[23] and also higher than HCN content in another mangrove fruits such as *Avicennia marina* 8.37 ppm.[24] Nevertheless, soaking in $CaCO_3$ solution can reduce HCN content in the *lindur* fruits by 67%. It is assumed that CN on HCN binds to Ca on $CaCO_3$. Tan et al.[25] conducted a study to reduce of HCN with CaO at high temperature. HCN and CaO react and produce $CaCN_2$, CO, and H_2.

5. CONCLUSION

The different soaking treatments could affect the nutritional (proximate composition and crude fiber) and antinutritional properties (tannin content and HCN content) of *lindur* fruit flour. The saturated salt solution soaking treatment was the best pretreatment to reduce antinutritional properties of *lindur* fruit flour. *Lindur* fruits flour with the low level of anti-nutritional components can be safe for human consumption.

The presence of antinutritional factors identified in this current study illustrated that these should not pose a problem to human health if the *lindur* fruit is properly processed. The pre-treatments with soaking can reduce the tannin and HCN content in the *lindur*

fruits flour. In view of the overall proximate composition, *lindur* fruit can be explored as an alternative source of food.

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