Nata de cincau: Pacitan’s black cincau (Mesona palustris BL) product innovation with respect to its physical, chemical and sensory characteristics

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Abstract. Black cincau (Mesona palustris BL) is a particularly abundant commodity in Pacitan, Indonesia. It has a higher fibre and antioxidant content compared to other varieties. Black cincau is potentially processed into nata de cincau in order to diversify food products in Pacitan. The aim of this study is to define the best formulation of nata de cincau, considering its physical (yield, thickness and texture) and chemical (water and crude fibre) characteristics. The best three formulations, based on physical and chemical characteristics, were chosen for sensory evaluation (colour, flavour, taste, texture and overall). Nata de cincau processing used various concentration of substrates (extract black cincau) (2.5%, 7.5% and 12.5%) and a variety of starter concentrations (5%, 10% and 15%) were added. Nata de cincau with 12.5% substrate concentration and 5%, 10% and 15% starter concentration were the best three formulations in terms of all physical and chemical characteristics. Sensory evaluation was performed on the best three formulations; 12.5% substrate concentration and 15% starter concentration was found to be the best formulation.

1. Introduction
Black cincau (Mesona palustris BL) is a plant that grows wild on hillsides and forests in Pacitan, Wonogiri and Ponorogo, which is known locally as Janggelan. Black cincau is a shrub with a height of 30–60 cm and grows at an altitude of 150–1,800 above sea level [1]. Black cincau is highly abundant in Pacitan. The success of cultivating black cincau in this region has not been followed by a good post-harvest process. The most common post-harvest problem is the lack of processed black cincau diversification. Normally, people only sell black cincau in the form of dried black cincau and gel [2]. Black cincau has a nutritional content of 122 kcal energy, 6 g protein, 1 g fat, 26 g carbohydrates, 17 mg vitamin C and 40% digestible ingredients [3]. Further, the antioxidant content of black cincau is higher compared to green cincau and the shrub cincau [4].

Nata is a processed food product which has the form of a gel, chewy texture and white-coloured [5]. According to some studies, nata can be made from coconut water [6], seaweed [7], pineapples [8], cassava [9] and melon with papaya [10]. Nata is an organic product that contains high fibre [11]. The ideal liquid substrate for Acetobacter xylinum in making nata has to contain at least carbohydrates, proteins and vitamins to provide a significant effect on growth and production of nata. Sodium, potassium, calcium, magnesium and iron have been found to be important nutrients in the formation of polysaccharides [12]. This study analyzes food products based on
Pacitan’s local wisdom: nata made from black cincau (nata de cincau), This might be the first report on the utilization of black cincau as raw material of nata production. The influence of substrate and starter culture concentration on physical and chemical characteristic as well as sensory attributes of nata de cincau was studied in this study.

2. Experimental
2.1. Sample preparation
Black cincau (Mesona palustris BL) was weighed to 2.5%, 7.5% and 12.5% (w/v). Black cincau was then boiled with one litre of water for five minutes at 100°C. The next process was the disposal of black cincau dregs, followed by obtaining the extract water. The water was boiled again, then 12.5% sugar, 2.5% ammonium sulphate and 1.5% citric acid were added to the boiling water. The extract water was poured into a tray, which was tightly closed. The extract water was left to stand for 24 hours at room temperature. Acetobacter xylinum obtained from local producer of nata de coco at Sragen City, Central Java was then inoculated into the extract water at 5%, 10% and 15% (v/v) concentration and then fermented for seven days at room temperature.

2.2. Analytical methods
The analysis of this research included physical parameters such as yield (using a gravimetric method) [13], thickness (with a Kenmaster Vernier caliper), texture (texture analyzer CT3) and chemical (water content: using gravimetric method; crude fibre content: employing a gravimetric method) [14]. The best three formulations were chosen from physical and chemical characteristics and were examined in terms of their organoleptic properties (colour, flavour, taste, texture and overall) with a 5-point scoring scale 1) Dislike, 2) Dislike moderately, 3) Neither like nor dislike, 4) Like moderately, 5) Like [15]. This sensory evaluation for nata de cincau involved a group of panellists, comprising 60 random untrained and semi-untrained panellist members.

2.3. Statistical analyses
Data physical and chemical tests were analyzed using two-way ANOVA to determine whether there were any differences in each treatment. If differences were noted, the test was then continued by using a Duncan Multiple Range Test (DMRT) at a significance level of α = 0.05.

3. Results and Discussion
3.1. Physical characteristics

| Substrate (%) | Yield (%) | Thickness (cm) | Texture (gram) |
|---------------|-----------|----------------|----------------|
| 2.5 | 5 | 42.04±1.41<sup>Aa</sup> | 0.60±0.02<sup>Aa</sup> | 521.33±0.02<sup>Aa</sup> |
| | 10 | 50.62±1.46<sup>Ab</sup> | 0.70±0.02<sup>Ab</sup> | 549.42±0.08<sup>Ab</sup> |
| | 15 | 59.26±1.63<sup>Ac</sup> | 0.79±0.02<sup>Ac</sup> | 560.58±0.04<sup>Ac</sup> |
| 7.5 | 5 | 52.96±1.34<sup>Ba</sup> | 0.78±0.03<sup>Ba</sup> | 571.33±0.03<sup>Ba</sup> |
| | 10 | 60.33±1.17<sup>Bb</sup> | 0.89±0.02<sup>Bb</sup> | 581.50±0.01<sup>Bb</sup> |
| | 15 | 68.48±1.16<sup>Bc</sup> | 1.00±0.02<sup>Bc</sup> | 588.58±0.04<sup>Bc</sup> |
| 12.5 | 5 | 61.77±1.47<sup>Ca</sup> | 0.91±0.01<sup>Ca</sup> | 600.33±0.02<sup>Ca</sup> |
| | 10 | 69.27±1.22<sup>Cb</sup> | 1.10±0.02<sup>Cb</sup> | 615.67±0.02<sup>Cb</sup> |
| | 15 | 76.86±1.20<sup>Cc</sup> | 1.30±0.02<sup>Cc</sup> | 634.17±0.03<sup>Cc</sup> |

Values within a column with different superscripts (A–C) denote significant differences from each other as affected by different substrate concentrations, p ≤ 0.05.
Values within a row with different superscripts (a–c) denote significant differences from each other as affected by different starter concentrations, p ≤ 0.05.
Yield is the percentage ratio between the final weight product and initial weight of the material in the form of substrate, water and nutrients (sugar, ammonium sulphate, pH) multiplied by 100%. Based on Table 1, substrate and starter concentrations were significantly different between samples. Results also showed that the percentage of yield increased with increasing substrate and starter concentration. The sample with the highest yield (76.86%) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. During fermentation, Acetobacter xylinum hydrolyzes sugar into polysaccharides or extracellular cellulose. A high yield indicates that Acetobacter xylinum was able to break down sugar to form nata. If the nutrients (carbon, nitrogen and minerals) and pH were not suitable for the fermentation process, the growth of Acetobacter xylinum would be inhibited. Appearance of nata de cincau samples are showed at Figure 1 and Figure 2.

3.1.2. Thickness
Based on Table 1, the thickness of nata is significantly influenced by substrate and starter culture. A strong correlation between factors and thickness were exhibited by Pearson correlation test, with the result of 0.918. Results showed that the thickness level increased with increasing substrate and starter concentration. The thickest sample (1.3 cm) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. Some factors that lead to increased thickness include substrate and starter concentration, nitrogen and carbon source, size of tray and duration of fermentation.

3.1.3. Texture
Texture of the samples was tested using a CT3 Texture Analyzer with Texture Profile Analysis (TPA) mode. Based on Table 1, substrate and starter concentration were significantly different between samples and there were interactions between factors. The interactions between factors were tested using a Pearson correlation test; the result of 0.704 is indicative of a strong correlation between them. Results also demonstrated that the texture level increased with increasing substrate and starter concentration. The most textured sample (634.17 g) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. The texture of nata is related to its crude fibre and water content. High crude fibre lead to nata’s texture becoming chewy, while high water content causes excessive amounts of water to enter the cellulose tissue and subsequently the texture becomes harder.

3.2. Chemical Characteristics
3.2.1. Water Content
Water content is very important in determining food shelf life because it affects physical and chemical properties, microbiological changes and enzymatic reactions. Based on Table 2, substrate and starter concentrations were found to be significantly different between samples. Results showed that the water content percentage increased with increasing substrate and starter
concentration. The sample with the highest percentage of water content (93.34%) was nata de *cincau* with a substrate concentration of 12.5% and starter concentration of 15%. The main factor that affected water content is sugar concentration. Water content percentage increased with increasing sugar addition. Sugar leads to several cavities in nata, where water will most likely be trapped [17].

**Table 2. Chemical characteristics data**

| Substrate (%) | Starter (%) | Water content (%) | Crude fiber (%) |
|---------------|-------------|-------------------|----------------|
| 2.5           | 5           | 83.19±0.62<sup>Aa</sup> | 1.53±0.07<sup>Aa</sup> |
|               | 10          | 85.23±0.52<sup>Ab</sup> | 1.76±0.07<sup>Ab</sup> |
|               | 15          | 87.27±0.67<sup>Ac</sup> | 1.98±0.09<sup>Ac</sup> |
| 7.5           | 5           | 86.20±0.72<sup>Ba</sup> | 1.86±0.07<sup>Ba</sup> |
|               | 10          | 88.33±0.56<sup>Bb</sup> | 2.07±0.08<sup>Bb</sup> |
|               | 15          | 90.38±0.59<sup>Bc</sup> | 2.26±0.06<sup>Bc</sup> |
| 12.5          | 5           | 89.31±0.66<sup>Ca</sup> | 2.14±0.04<sup>Ca</sup> |
|               | 10          | 91.36±0.57<sup>Cb</sup> | 2.37±0.08<sup>Cb</sup> |
|               | 15          | 93.34±0.66<sup>Cc</sup> | 2.57±0.07<sup>Cc</sup> |

Values within a column with different superscripts (A–C) denote significant differences from each other as affected by different substrate concentrations, p ≤ 0.05.

Values within a row with different superscripts (a–c) denote significant differences from each other, as affected by different starter concentrations, p ≤ 0.05.

3.2.2. **Crude Fibre Content**

Crude fibre is a part of food that cannot be hydrolyzed by acids and bases. The acid used to test for crude fibre is sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and the base is sodium hydroxide (NaOH). Based on Table, substrate and starter concentrations were significantly different between samples. Results showed that the crude fibre percentage increased with increasing substrate and starter concentration. The sample with the highest percentage of crude fibre content (2.54%) was nata de *cincau* with a substrate concentration of 12.5% and starter concentration of 15%. High fibre in nata is affected by bacterial activity, while such activity is affected by sugar as the source of carbon in the fermentation process. *Acetobacter xylinum* converts sugar into cellulose to produce fibre [23].

The best three formulations based on physical and chemical test results were then evaluated using sensory tests. These three best formulations were nata de *cincau* with a substrate concentration of 12.5% and starter concentrations of 5%, 10% and 15%.

3.3. **Sensory Characteristics**

An organoleptic test was carried out by using a food-preference test, with in this study, the higher score implying better quality. Data regarding the sensory characteristics test (colour, flavour, taste, texture, overall) are presented in Table 3.

**Table 3. Sensory characteristics data**

| Substrate (%) | Starter (%) | Colour   | Flavour   | Taste    | Texture  | Overall  |
|---------------|-------------|----------|-----------|----------|----------|----------|
| 2.5           | 5           | 2.80±0.8<sup>a</sup> | 2.55±0.8<sup>a</sup> | 2.20±0.7<sup>a</sup> | 2.35±0.6<sup>a</sup> | 2.48±0.4<sup>a</sup> |
|               | 10          | 3.06±0.9<sup>b</sup> | 3.11±0.8<sup>b</sup> | 2.64±0.7<sup>b</sup> | 2.64±0.6<sup>a</sup> | 2.86±0.3<sup>b</sup> |
|               | 15          | 3.61±0.9<sup>c</sup> | 3.69±0.7<sup>c</sup> | 3.35±0.7<sup>c</sup> | 3.30±0.8<sup>b</sup> | 3.49±0.4<sup>c</sup> |

Values within a row with different superscripts (a–c) denote significant differences from each other, as affected by different starter concentrations, p ≤ 0.05.
3.3.1. Colour
The colour of a product is an important factor that shows the first impression of sensory characteristics seen by panellists. Determining food’s initial quality often begins with its colour [24]. Results showed that the colour preference score increased with increasing bacterial concentration. The sample with the highest score in colour preference (3.61 or like moderately) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. Common nata of good quality should be transparent white, slippery and rather shiny, but nata de cincau is blackish brown due to the addition of black cincau [25]. Black cincau has flavonoid compounds, which cause a distinctive dark colour that will change nata or any other products’ colour to be rather dark [26, 27].

3.3.2. Flavour
Flavour is one of the quality parameters in food products that determines whether or not a product is good, using the sense of smell [25]. Based on Table 3, the sample with a starter concentration of 15% was significantly different from the others. Results showed that the flavour preference score increased with increasing bacterial concentration. The sample with the highest score in flavour preference (3.69 or like moderately) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. Human senses can only detect at least four smells: sour, sweet, rancid, and charred/burnt, although the flavor of nata de cincau is neutral with a little sour [28]. The sour aroma can be removed by soaking nata in fresh water and boiling it for ten minutes at 100 °C during the harvesting process after fermentation. The boiling needs to be repeated until the smell has completely vanished [29, 25].

3.3.3. Taste
Results demonstrated that the taste preferences score increased with increasing bacterial concentration. The sample with the highest taste preference (3.35 or neither like nor dislike) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. Nata de cincau was tasteless with a rather sour and slightly sweet. The sour taste of nata is produced by glucose, which is hydrolyzed into cellulose in the fermentation media. While nata is being soaked and boiled, the osmosis process releases the sour taste and nata becomes tasteless [22].

3.3.4. Texture
Texture is one of the organoleptic parameters that determines product quality. The human senses used in observing the texture are usually fingers, teeth and palate. A good nata appears in the form of a gel, with its texture tending to be chewy, tender and transparently white in colour [30, 29, 31]. Results illustrated that the texture preferences score increased with increasing bacterial concentration. The sample with the highest texture preference (3.3 or neither like nor dislike) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%. This result indicated that nata de cincau had an acceptable texture according to SNI 01-4317-1996, which states that the texture of a nata must be normal. The texture parameter is affected by several factors such as water content, duration of fermentation and crude fibre content [30, 29, 19].

3.3.5. Overall
Results demonstrated that the overall preferences score increased with increasing bacterial concentration. The sample with the highest overall preference (3.49 or neither like nor dislike) was nata de cincau with a substrate concentration of 12.5% and starter concentration of 15%.

4. Conclusion
In sum, Pacitan’s black cincau (Mesona palustris BL) can be processed into nata de cincau. The best three samples in terms of physical and chemical characteristics were nata de cincau with a substrate concentration of 12.5% and starter concentrations of 5%, 10% and 15%. These samples
were then subjected to a sensory evaluation using an organoleptic test, considering their performance with respect to colour, aroma, taste, texture and overall. The sample with a substrate concentration of 12.5% and starter concentration of 15% was found to be the best formulation of nata de cincau.

5. References
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