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by Miksusanti Salbi

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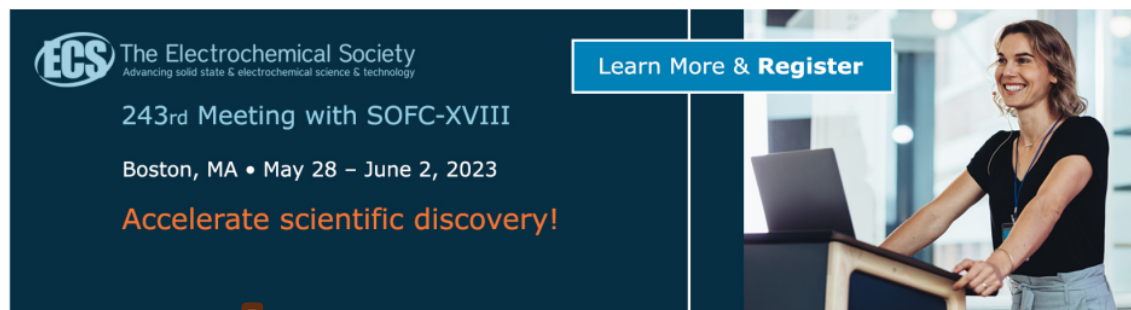
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Determination of quality parameters in nata de cocolawak as hepatoprotector functional food

Indah Solihah¹, Miksusanti^{2*}, Rennie Puspa Novita¹, Sandy Yoga Ramadhan¹

¹ Department of Pharmacy, Faculty of Mathematics and Natural Sciences, Sriwijaya University

² Department of Chemistry, Faculty of Mathematics and Natural Sciences, Sriwijaya University

Correspondent author: *miksusanti@gmail.com

Abstract. One of the hepatotoxic condition is caused when consuming large amounts of drugs induced liver injury for a long period of time, such as in TB (tuberculosis) treatment [1]. One of the solutions to prevent the occurrence of hepatotoxicity symptoms in TB patients is by consuming temulawak (*Curcuma xanthorriza* Roxb.) supplements [2]. Temulawak supplement was given in the form of dessert nata de cocolawak to increase patient interest in consuming it because it is served in a better taste. Parameters product quality of food was determined based on SNI (Indonesian National Standard). This research was conducted in order to increase the economics and environment value of nata de coco products as a functional food. Based on the measurement results, the quality of nata de cocolawak products meets most of the SNI parameters for nata de coco products. Nata de cocolawak products have 9.56% of sugar, 2.11% of crude fiber, level of mineral content are 3.63 mg/Kg of iron, 58.25 mg/Kg of calcium, and 25.73 mg/Kg of zinc, level of metal contaminant are 0.005 ppm of Pb, 0.212 ppm of Cu, and negative levels for As. The Total plate count (TPC) on the product to measure microbial contamination is $5.5 \cdot 10^2$ CFU/g. Curcumin content as an active compound in the product is 1.306mg/g of sample. The treatment group had smaller ALT, AST, and ALP values than the placebo group. But statistically, these values did not differ significantly ($p > 0.05$).

1. Introduction

Liver dysfunction is still a major health problem in both developed and developing countries. Indonesia is a country with a high endemic rating of liver disease [3]. The incidence of liver damage is very high, starting from damage that is not fixed but can last a long time [4]. One of the causes of liver damage is drugs [3]. In the United States alone there are around 2000 cases of acute liver failure that occur every year and more than 50% are caused by drugs [5]. Drugs that are said to be hepatotoxic are drugs that can induce liver damage or are usually called drug-induced liver injury [6]. Liver damage-inducing drugs are increasingly recognized as a cause of acute and chronic liver disease [7].

The majority of tuberculosis medicines are drugs induced liver injury. Rifampicin can cause obstructive jaundice. Isoniazid can cause liver parenchymal damage both quickly and slowly, cause granuloma of liver cells, and induce fat infiltration in liver cells. While pyrazinamide can cause rapid liver parenchymal damage [8].

One solution to prevent the onset of symptoms of hepatotoxicity in tuberculosis patients is the administration of temulawak supplements. Temulawak is an Indonesian native plant that has been shown to have hepatoprotective activity in cisplatin-induced hepatotoxic mice. The mechanism of



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temulawak as a hepatoprotector by reducing serum enzyme levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), and γ -glutamyl transferase (GGT) [9]. In addition, temulawak can also prevent liver adipose (liver fat degeneration) which can cause irreversible damage to liver function [10]. The main active compound of Temulawak is curcumin. Curcumin has hepatoprotector activity as an antioxidant with scavenging radical superoxides ion and increasing endogenous antioxidant concentration [11,12]. Temulawak supplements can be given in the form of a nata de cocolawak dessert. The provision of supplements in the form of dessert has the aim of increasing the patient's interest in consuming it because it is served in a better taste.

2. Experimental Section

2.1. Materials and Chemicals

The materials used in this study were nata de cocolawak, curcumin, peptone dilution fluids (PDF), plate count agar (PCA), acetone, ethanol, aquadest, sodium hydroxide, hydrochloric acid, sulfuric acid, ALT kit reagent, AST kit reagent, ALP kit reagent.

2.2. Sugar Content

Sugar content was measure based on SNI 01-2891-1992 at the 3rd point. The sample was tested at Chemistry and Microbiology Agricultural Laboratory, Sriwijaya University.

2.3. Crude fiber content

Crude fiber content was measure based on SNI 01-4317-1996 at 5.6 points. The sample was tested at Chemistry and Microbiology Agricultural Laboratory, Sriwijaya University.

2.4. Mineral content

Total Fe, Ca, and Zn content on nata de cocolawak product was determined with a wet destructive method using AAS (Atomic Absorption Spectroscopy) [13]. The sample was tested in the Integrated Testing Laboratory, Faculty of Mathematics and Natural Science Sriwijaya University.

2.5. Metal contaminant content

Total Pb, Cu, and As content on nata de cocolawak products were determined with a wet destructive method using AAS (Atomic Absorption Spectroscopy) [13]. The sample was tested in the Health Laboratory of Palembang, South Sumatera.

2.6. Microbial contamination

A total of 5 tubes were filled with 9 mL peptone dilution fluids (PDF). The homogenization results from the preparation of the sample were plated by 1 mL dilution into the tube containing the first PDF diluent until 10^{-2} dilution was obtained and shaken to homogeneous, further diluting until reached the 10^{-6} dilution. One mL of each dilution was poured into a petri dish and duplicated, then poured 15 - 20 mL of medium plate count agar (PCA). Petri dish was shaken and treated in such a way until the suspension spread evenly. To determine the sterility of the media and diluent, a control test (blank) was made. After the media was solidified, the petri dish was incubated at 35 - 37 °C for 24 - 48 hours with the upside down position. The number of growing colonies was observed and calculated [14].

2.7. Curcumin content of the product

Determination of curcumin content on nata de cocolawak product according to a method described previously [15].

2.8. Hepatoprotector Test Procedure

This study was a clinical experimental study with placebo-controlled group design, which was registered in Health Research Review Committee Mohammad Hoesin Central General Hospital and Faculty of Medicine Sriwijaya University, and the registry number is 063/kepkrsmhfkunsri/2019. The sample was randomly selected with 36 healthy women, divided into 2 treatment groups. Respondents

were selected by inclusion criteria, which are women, adults (17-40 years), healthy, normal liver physiological values, and not taking drugs induced hepatotoxic. All respondents received the same nutritional intake during the experiment. The independent variable in this study was giving a 100g product (nata de cocolawak for treatment group, and nata de coco for the placebo group) 3 times a day for 30 days. The dependent variable in this study was the level of ALT, AST, GGT and ALP of the test subjects. Determination level of AST, ALT, and ALP serum according to a method described previously [16] in Sriwijaya University Health Center Clinic.

3. Result and Discussion

The quality of processed food will be guaranteed if it can meet the parameters set by the NSA (National Standardization Agency). Every parameter set by the NSA applies nationally through SNI (Indonesian National Standard). So that for nata de cocolawak product, to be able to assess this product quality or not, it can be compared with the SNI parameters for similar products, such as nata de coco. The results show in table 1.

Table 1. Nata de cocolawak quality measurement results

Parameters	Result	Reference ^a	RDA (mg per day per person) ^b
Sugar Content (%)	9.56	Min. 15	309-375. 10 ³
Crude Fiber content(%)	2.11	Max. 4.5	32-38. 10 ³
Iron content (mg/Kg)	3.63	-	13-26
Calcium content (mg/Kg)	58.25	-	1100
Zinc content (mg/Kg)	25.73	-	10-13
Lead contamination (ppm)	0.005	Max. 0.2	-
Arsenic contamination (ppm)	Negative	Max. 0.1	-
Copper contamination (ppm)	0.212	Max. 2	-
Total plate count (cfu/g)	5.5.10 ²	Max. 1.10 ⁴	-

Source: a. SNI 01-4317-1996 b. Indonesian Health Ministry regulatory number 75 the year 2013 for men/women adults age 19-29th

Based on the data in table 1, nata de cocolawak product meets the standards of SNI, except for the total amount of sugar that is less than the minimum SNI standard. The taste of nata de cocolawak product is less sweet and tends to be slightly bitter due to the addition of dried temulawak rhizomes in the process of making nata de cocolawak. Beside that, this product has several minerals, such as iron, calcium, and zinc that importance for health. Iron has several vital functions in the body. It serves as a carrier of oxygen to the tissues from the lungs by red blood cell hemoglobin, as a transport medium for electrons within cells, and as an integrated part of important enzyme systems in various tissues [17]. Zinc plays a central role in the immune system, affecting a number of aspects of cellular and humoral immunity [18]. Calcium plays a central role in bone formation and calcium ion plays a role in many metabolic processes.

The level of curcumin in nata de cocolawak product is determined to find out how much curcumin is extracted into the product. The determination of the level of curcumin begins with measuring the curcumin calibration curve. The results of the subsequent absorbance measurements are linear regression so that the standard curve equation is $y = 0.1006x + 0.0606$ as seen in Figure 2.

Based on the equation of the standard curve, the curcumin content of nata de cocolawak product is calculated as a 1.306 mg/g sample. The extracted curcumin content in the sample is only about 0.7% of the curcumin content contained in the dried temulawak rhizome. This is because curcumin is semipolar so it will be difficult to extract in a polar solvent such as water.

The study of hepatoprotector effect was done by measuring ALT, AST, and ALP level enzyme value of the respondent. Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are two types of enzymes produced by liver cells. Both of these enzymes are used as indicators for examining liver function, where levels will increase in the blood when liver cells are damaged. Examination of alkaline phosphatase (ALP) provides information on the presence or absence of a

process of damage in the hepatobiliary system. If bile or liver damage occurs, the liver will increase ALP synthesis so that the amount of ALP in the blood increases [19].



Figure 1. Comparison of product with (1) and without temulawak (2)

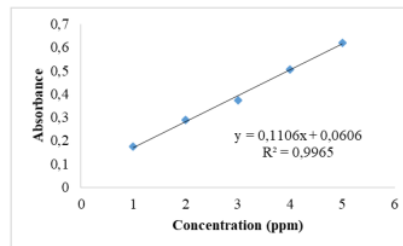


Figure 2. Calibration curve of curcumin

Respondents were grouped into 2 groups, namely the placebo group who consumed nata de coco, and the treatment group who consumed nata de cocolawak. They received 100 g product 3 times a day for 30 days. This is considered that the level of curcumin contained in the product is very small, so to optimize the effect, we give a lot of product to be consumed. Each respondent received the same nutritional intake to prevent the occurrence of bias in the measurement results later. The determination of ALT, AST, and ALP levels was carried out on day 30 by spectrophotometry. The result can be seen in table 2.

Table 2. Levels enzyme value of ALT, AST, and ALP of respondent

Parameters	Differential value (treatment – placebo) (mean±SD)	p-value
ALT (U/L)	-2.53±0.42	0.724
AST (U/L)	-0.15±0.31	0.650
ALP (U/L)	-8.28±5.18	0.463

Based on the results in table 2 it can be seen that the treatment group had smaller ALT, AST, and ALP values than the placebo group. But statistically, these values did not differ significantly ($p > 0.05$). So it can be concluded that consuming nata de cocolawak did not significantly influence the values of hepatic enzymes. This happens because the level of curcumin contained in the product is too small. Actually, nata de cocolawak has the potential to be used as a hepatoprotector supplement by increasing levels of curcumin in its products.

4. Conclusion

Nata de cocolawak meet most of SNI parameters and have curcumin content 1.306 mg/g sample. The treatment group had smaller ALT, AST, and ALP values than the placebo group, but not significant ($p > 0.05$). This product potential to be used as a hepatoprotector supplement by increasing levels of curcumin.

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