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The effect of *Sargassum binderi* alginate extract addition on the nutritional content of full cream Mung Bean Milk and its consumer acceptance

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Abstracts

Corresponding author: rais.nurlatifah@walisongo. ac.id Received : 20 March 2021 Revised : 20 May 2021 Accepted : 1 Juni 2021 Milk is one of the important nutritional components that consumed by children and adults. Mung Bean (*Vigna radiata L.*) contains high nutrition and functional compounds, as well as high nutrients content that the body needs. Mung Bean milk is processed into full cream Mung Bean milk with alginate addition. Variations of alginate addition are 1%, 1.5%, 2%, 2.5% and 3% (w/v). Characterization of full cream mung Bean milk is analysis of the viscosity, density, nutritional, total fiber content, and organoleptic analysis. Alginate is a heteropolysaccharide that has two monomers. The color of alginate is brown. The result of research in density and viscosity of the alginate are 1.43 g/mL and 7.42x10⁻³ kg m⁻¹ s⁻¹. The best composition for addition alginate to full cream Mung Bean milk is 3% (w/v). The result of the best full cream Mung Bean milk of carbohydrate, protein, fat, and total fiber content is 98 mg carbohydrates/mL milk; 0.97% in 5 mL milk; 0.32 mg fat/mL milk and 13.1 mg/mL milk. Organoleptic analysis of 3% (w/v) addition alginate in full cream Mung Bean milk is like category.

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Keywords: Mung Bean milk, Sargassum binderi, alginate

1. Introduction

Milk is one of the important nutritional components that the body needs [1]. The need for milk is the primary need at this time [18].

Milks are consumed by children and adult. Vegetable milk is a product that is also popular in society [2][19].

This study used Mung Beans milk for its experiments [3]. Mung Beans contain high nutrition and functional compounds that the body needs [4]. Mung Bean (Vigna radiata L.) is a legume plant and the third most widely cultivated after Soy Beans and peanuts [13]. These compounds include polyphenols and beta carotene [5]. Besides that, Mung Beans contain high antioxidant compounds and are able to ward off free radicals [6]. Mung Beans are source of vegetable protein [7]. Mung Beans contain protein, calories, fat, calcium, vitamin A, iron, vitamin C and vitamin B1 per gram of Mung Bean seeds [8], as well as vegetable protein, bioactive compounds, and minerals [20]. These advantages include more dry resistance, more resistance to pests and can grow in less fertile soils [9]. Besides that, Mung Beans have a short harvest life of 55-60 days [10].

Mung Bean milk is a kind of vegetable milk that can be consumed by people with lactose intolerance [3]. Mung Bean milk is also an alternative as a non-dairy protein source [16]. Mung Bean milk can be processed into full cream Mung Bean milk. The nutritional value of vegetable milk processing can also be enhanced with alginate. Alginate is extracted from Sargassum binderi [11]. Sargassum binderi is a type of brown seaweed [14]. In Indonesia, seaweed is a commodity that is abundant and easy to find in coastal waters [15]. Alginate is a heteropolysaccharide compound group that has two monomers [17]. The monomers are chain mannuronic monomers acid (poly-Dmannuronic acid) and guluronic acid (poly-Lguluronic acid) [12].

In this study, alginate was mixed with Mung Bean full cream milk to produce Mung Bean milk with higher nutrition. The effect of adding alginate extract was observed to determine its nutritional content.

2. Experiments Procedure

Processing of Sargassum binderi Powder

Sargassum binderi was taken from Gunung Kidul Beach, Yogyakarta. The processing of Sargassum binderi was started with washing it using distilled water. Sargassum binderi was then dried in the oven for 24 hours at 60oC. Next, the Sargassum binderi was freezed dry again to remove any remaining water. The dried *Sargassum binderi* was then blended and then filtered with a 180mesh filter. *Sargassum binderi* powder was then stored for the next extraction process.

Alginate Extraction from Sargassum binderi

Sargassum binderi powder soaked in 2% CaCl2 for three hours then washed with aquadest. The samples were then immersed in formaldehyde solution for 2 hours and washed again with aquabidest 3 times. Next, the sample was extracted with 3% Na2CO3 1 M and 0.5 grams of EDTA at pH 11. Then the sample was filtered using cloth and deposited using ethanol as sodium salt. The precipitate was separated by centrifugation and dried in an oven at 60oC.

The Process of Making Edible Film

Edible films were made with a composite mixture of hydrocolloid and glycerol. Alginate was used for homogenization of liquid and solid phases in milk. Its hydrocolloid used was alginate extract from macro algae *Sargassum binderi*. The alginate concentrations used were 1%, 1.5%, 2%, 2.5% and 3% (w/v). Alginate and glycerol were mixed and stirred until an emulsion is formed. Then the emulsion formed was put in full cream Mung Bean milk.

Pasteurization of Full Cream Mung Bean Milk

The milk sample was put in a pasteurization pan. Pasteurization was carried out at a temperature of 95oC for 3-4 minutes. The pasteurization process was repeated three times. Then the sample was cooled in a coldwater bath with continuous running water. The cooling process was carried out until the milk sample was at a temperature of 25-30oC.

Analysis of The Viscosity of Full Cream Mung Bean Milk

The viscosity of the milk samples was measured with an Ostwald viscometer. 10 mL of the milk sample was inserted into a viscometer. The milk sample was sucked over the upper line. Then the suction device was released and the stopwatch was activated when the liquid coincides with the top line of the tool. The stopwatch was then turned off when the liquid reaches the bottom line. The measurement then was repeated five times.

Density Analysis of Full Cream Mung Bean Milk

A Pycnometer was cleaned with acetone. Then the pycnometer was dried with a tissue and hair dryer. The blank pycnometer was weighed by mass. Furthermore, the pycnometer containing water was weighed by its mass. Then the pycnometer containing the milk sample was weighed. The measurement then was repeated five times.

Analysis of Nutritional Full Cream Mung Bean Milk

Nutritional analysis includes analysis of carbohydrate content, protein content, and fat content. Nutritional analysis uses the Indonesian National Standard (SNI) procedure for testing beverages.

Analysis of Total Fiber Content in Full Cream Mung Bean Milk

Ten (10) mL of sample was weighed and put into Erlenmeyer and added 25 mL of 0.1 M sodium phosphate buffer pH 6. The sample was added with 100 mg enziplex, and then the Erlenmeyer was covered with aluminum foil and incubated for 15 minutes at 80oC. The sample was allowed to cool and 20 mL of distilled water was added and the pH was adjusted to 1.5 by adding 4 M HCl. Then the sample was added with 100 mg of enzyme and incubated for 60 minutes at 40oC. The sample was added with 20 mL of distilled water and the pH was adjusted to 6.8 with 1% NaOH. Furthermore, the sample was added with 100 mg enziplex. Enzyplex is a drug that contains amylase, protease, deoxycholid acid, vitamin B1, vitamin B2, vitamin B6, niacinamide, and calcium panthothenate. Then the samples were put and incubated for 60 minutes at 40o C. Samples were then was filtered

with a Buchner funnel. Then the residue was washed with 2x10 mL of distilled water, and the sample was dried at 105oC for 4 hours. The samples were weighed to constant weight. Then the filtrate was diluted to 100 mL and 80 mL 95% ethanol added. Next the sample was allowed to settle for 1 hour. Then the solution was filtered with a buchner, and the sample was washed with 2x20 mL 95% ethanol and 2x10 mL acetone. The sample was dried at 105oC overnight, and then the sample was weighed until reaching a constant weight.

Organoleptic Analysis of Full Cream Mung Bean Milk

Organoleptic analysis used the hedonic scale method. The test criteria included aroma, taste, texture, colour and viscosity. The sample was presented homogeneously to the public. The assessment results were expressed in a hedonic scale starting from 1 (dislike), 2 (slightly dislike), 3 (normal), 4 (like) and 5 (really like). Each respondent filled in the assessment list. There were 35 respondents participated in this research.

The hedonic scale was transformed into a numeric scale with the numbers increasing according to the level of preference and then the results of statistical analysis were performed on the numeric scale. The level of preference was determined by the average result of each panelist at the 95% level of confidence (Stand & National 1991):

$$P(\bar{U} - 1,96 \, Sn1/2 < \mu < \bar{U} + 1,96 \, Sn1/2)$$
$$\bar{U} = \frac{\sum_{i=1}^{i=n} U_i}{n}$$
$$S^2 = \frac{\sum_{i=1}^{i=n} (U_i - U)}{n}$$
(1)

where n is the number of panelists, \overline{U} is average quality value, Ui is the quality value of the panelists to i, where i = 1 to i = n, S is standard deviation of quality values, S2 is diversity of quality values; 1,96 is the coefficient of standard deviation at the 95% confidence level.

3. Result and Discussion

Alginate Characterization of Sargassum binderi

Sargassum binderi in this study was taken from the south coast of Gunung Kidul. Seaweed samples were taken in March 2019. The coordinates of the sampling cite are 650 32' 24" south latitude and 890 21' 43" east longitude, which are located around the main gate of the south coast of Gunung Kidul. Brown seaweed or Sargassum binderi can be found in this place.



Figure 1. Sayang Heulang beach (Latifah, 2017)



Figure 2. Sargassum binderi (Latifah, 2017)

Then the *Sargassum binderi* was extracted using CaCl2. The color of alginate is

brown. The density and viscosity of the alginate are 1.43 g/mL dan 7.42x10-3 kgm-1s-1.

Density Analysis of Full Cream Mung Bean Milk

Full cream Mung Bean milk with various alginate compositions, the density was determined by using a pycnometer. Density is a measurement of the mass per unit volume. The higher the density of all object so the greater the mass per volume. The following is the density data of full cream Mung Bean milk.

Table 1.	Density	data	from	full	cream	Mung	Bean
milk with various alginate compositions							

Density (g/mL)
1,054
1,125
1,234
1,289
1,347
1,654

Based on table 1, it shows that full cream Mung Bean milk increases its density along with the addition of alginate to milk. This indicates that there was an alginate emulsion in the milk. The increase in density shows the higher the thickness of the Mung Bean milk. Mung Bean Milk contains hydroxy groups which cause alginate attachment to water molecules in the Mung Bean milk.

Viscosity Analysis of Full Cream Mung Bean Milk

Full Cream Mung Bean Milk with variations of alginate addition was determined by the viscosity of the Ostwald viscometer. Figure 2 shows an increase in the viscosity value along with the addition of alginate. The increase in viscosity was due to the emulsion which the interaction between alginate and water formed a colloid system. There is an increased Van der Waals interaction so that the interaction between carbohydrate molecules and alginate increases as well. Alginates which are non-polar interact with carbohydrates that have long chains that are non-polar as well. In the alginate *Sargassum binderi* contains a carboxylate group which acts as an emulsifier.

The emulsifier functions as a surface tension lowering so that the components can unite where the two are connected by a polar group and a non-polar group on the emulsifier. The increase in viscosity value has an impact on the resulting milk products, namely milk that has a high viscosity value has a thicker milk texture.



Figure 3. Graph of the viscosity of full cream Mung Bean milk with the addition of alginate

Analysis of Nutritional Full Cream Mung Bean Milk

1. Analysis of carbohydrate content

The carbohydrate content of full cream Mung Bean milk with the addition of alginate was analyzed using the Luff Schroll's method. Here are the results.



Figure 4. Graph of the effect addition alginate to full cream Mung Bean milk on carbohydrate content

Figure 3 shows an increase in carbohydrate levels along with the addition of alginate. The addition of 3% (w/v) alginate showed the highest carbohydrate content, namely 98 mg carbohydrates/mL milk. The increase in carbohydrate levels is influenced by the addition of alginate to milk. Where alginate is a polysaccharida of carbohydrate. This has an impact on increasing levels of carbohydrates contained in milk, namely alginate contributing as a sugar reducing agent with monosaccharides found in full cream Mung Bean milk.

2. Analysis of protein content

Protein content determination was carried out using the Kjeldahl method. The principle of this method determines the total nitrogen content in the sample. Following are the results of the analysis.



Figure 5. Graph the effect of alginate addition on milk protein content full cream Mung Bean milk

Figure 4 shows an increase in protein levels with the addition of alginate to milk. The addition of 3% alginate has the highest protein content, namely 0.97% (w/v). The carbonate groups in the alginate form aggregates that cause new bonds to form in milk. This affects the structural changes in milk which have an impact on increasing the protein content in milk

3. Analysis of fat content

Analysis of fat content in milk was analyzed using gravimetric method. Following are the results of the analysis.



Figure 6. Weight of milk fat with variations in the addition of alginate

Figure 5 shows that the content of fat nutrients in full cream Mung Bean milk with the addition of alginate is lower than without the addition of alginate. Alginate can reduce the fat content in milk. Where the addition of alginate in the form of edible film mixed with glycerol forms a hydrogen bond between the carboxylate group of the alginate and the hydroxide group of glycerol. This interaction causes the molecule to be non-polar which can be extracted by nonpolar solvents as well as in hexane. So that the fat content produced from milk with the addition of alginate is decreases.

Total Fiber Content of Full Cream Mung Bean Milk

Fiber is a polysaccharide that has β glucoside bonds. The fiber were not degraded by the α -amylase and stomach acid enzymes. The addition of pH 6 sodium phosphate buffer stabilizes the alpha amylase enzyme. The purpose of addition alpha amylase from the enzyplex and heating breaks the starch by gelatinizing it. Setting the pH to 1.5 conditions for maximum pepsin enzyme activity. Adjusting the pH to 6.8 aims for maximum pancreatin enzyme activity. From the research, the fiber content in full cream Mung Bean milk with the addition of alginate is presented in the table below.

Table 2 shows the highest fiber content, namely in full cream Mung Bean milk with the addition of 3% alginate. Alginate can increase total dietary fiber content in milk. This shows that there is more fiber which can be digested by human digestive enzymes and can be used as a source of energy. A beverage product, alginate is a source of fiber that is easily soluble in water. When dissolved in water, sodium alginate forms a mesh-like lattice that is able to firmly bind many water molecules. This causes the fiber content in brown rice milk to increase.

Table 2. Total fiber content on full cream Mung Bean

 milk

Full cream Mung Bean milk	Mass of total fiber per mL of milk	Mass of crude fiber	mass of food fiber	
No added alginate	7.1	2.4	4.7	
1%	9.6	3.8	5.8	
1.5%,	10.3	3.9	6.4	
2%,	11	4.1	6.9	
2.5%	11.9	4.3	7.6	
3%	13.1	4.9	8.2	

Organoleptic Analysis of Full Cream Mung Bean Milk

Organoleptic analysis used the hedonic test (preference test). In the preference test carried out in this study, 35 untrained panelists were used. The samples tested were full cream Mung Bean milk with varying concentrations of the addition of alginate and the addition of 6% sugar in each sample.

Panelists were asked for their responses through a questionnaire attached to the appendix, after tasting a random sample of brown rice milk. The smallest rating scale describes dislike while the largest rating scale describes the panelist's liking. Scale 1 means dislike, scale 2 means somewhat disliked, scale 3 means normal, scale 4 means like and 5 means very like. The results of the responses from the panelists were then analyzed statistically.

Full cream Mung Bean milk with the addition of 3% alginate is the most preferred milk by the public. The resulting milk texture is suitable for the taste from children to adults.

Criteria	Sample A		Sample	B	Sample C	
	Score	Criteria	Score	Criteria	Score	Criteria
Taste	$2,42 < \mu < 3,35$	Little like	3,64 < µ < 4,27	Like	3,76 < µ < 4,27	Like
smell	264 < µ < 3,24	Little like	3,82 < µ < 4,25	Like	3,79 < µ < 4,32	Like
Color	2,72 < µ < 2,89	Little like	3,49 < µ < 3,91	Like	3,87 < µ < 4,31	Like
Viscosity	2,43 < µ < 2,89	Little like	3,68 < µ < 4,23	Like	4,23 < µ < 4,51	Like

Table 3. Hedonic test result if full cream Mung Bean milk

*Note: Sample A : Full cream Mung Bean milk+2% alginate; Sample B : Full cream Mung Bean milk+2.5 alginate; Sample C : Full cream Mung Bean milk+3% alginate

4. Conclusion

Full cream Mung Bean milk with the addition of 3% alginate is the most preferred milk by the public. Nutritional analysis of carbohydrate, protein, and fat content was 98 mg carbohydrates/mL milk , 0.97% (w/v), 0,3 mg fat/mL milk.

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