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Development of instant jelly powder from elephant foot yam (*Amorphophallus oncophyllus*) and roselle (*Hibiscus sabdariffa* L.) as a functional food for diabetes mellitus patients

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Abstract

Elephant foot yam (*Amorphophallus oncophyllus*) is one of Indonesia's most widely grown agricultural commodities. It has a myriad of benefits, especially as a functional food for people with diabetes mellitus. Roselle (*Hibiscus sabdariffa* L.) is a flowering herb rich in polyphenols and anthocyanins with antioxidant and antidiabetic potentials. Therefore, the aim of this study was to create a functional food that is suitable for diabetes mellitus patients. In this study, a functional food product in the form of instant jelly powder was developed from the yam and roselle. Glucomannan extract, obtained from the yam flour by leaching, was tested using a Fourier-transformed infrared (FTIR) spectrophotometer (qualitative) and determined the content. Three formulas with different amounts of glucomannan and iota carrageenan were developed to determine the most optimized formulation. The most optimized formulation was determined based on organoleptic characteristics and the results of the gel strength and hardness, total phenolic content (TPC), and antioxidant activity tests using the ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) assay. Glucomannan extraction produced a 92.40% yield, with 46.32% glucomannan content. The spectrophotometric analysis indicated the presence of glucomannan in the extract, and further tests showed it increased with the decrease in gel strength and hardness. It was found that formula I produced the best jelly texture with a total phenolic content of 0.30% GAE (formula II 0.13% GAE; formula III 0.27% GAE) and an ABTS radical scavenging activity of 90.51% (II: 73.49%; III: 88.16%). In conclusion, formula I containing 6.35 g of carrageenan, 2.12 g glucomannan, 1.5 g roselle, 0.03 g citric acid, and 0.003 g sucralose has the best composition to create the most elastic and firm jelly texture with the highest phenolic content and radical scavenging activity.

1. Introduction

Diabetes mellitus is a chronic metabolic disorder characterized by an elevated glucose level in the blood, which can cause damage to the heart, blood vessels, eyes, kidneys, and nerves (World Health Organization [WHO], 2021). According to the International Diabetes Federation, by 2021, the number of people with diabetes will have reached 537 million worldwide-90 million in Southeast Asian countries-with a mortality rate of 6.7 million and around 541 million adults being at high risk of type 2 diabetes (International Diabetes Federation [IDF], 2021). Also, 2014 recorded that 8.5% of adults aged 18 years and over had diabetes, one of the direct causes of 1.6 million deaths. Indonesia currently ranks sixth in the world with 10.3 million people with diabetes (IDF, 2015). In 2013, type 2 diabetes, commonly found in adults, accounted for 90% of the total diabetes cases (Kemenkes RI, 2013). To control these cases, pharmacological therapies with antidiabetic drugs and non-pharmacological treatments with lifestyle improvement have been introduced. Because dietary habits and lifestyle greatly determine the success of the management and prevention of type 2 diabetes, a nutritional approach is vastly implemented to create an effective diet pattern (Forouhi *et al*., 2018)

Amorphophallus oncophyllus, known as elephant

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foot yam (English) or porang or iles-iles (Indonesian), grows in soils with high fertility, loose structure, and relatively low acidity (Sumarwoto and Maryana, 2011). It mainly contains a high concentration of glucomannan (49-60%), a water-soluble fiber. Glucomannan simultaneously improves metabolic control and reduces cardiovascular risk factors in people at high risk of diabetes (Vuksan *et al*., 1999) as it acts on glucose homeostasis and lipid metabolism (Susanti, 2014) and can inhibit the rate of glucose absorption in the digestive system. In the health sector, glucomannan can be used to treat diabetes, obesity, and hypercholesterolemia and reduce constipation (Chen *et al*., 2019). In previous research, giving effervescent tablets containing elephant foot yam at 0.18 g/200 g to test animals for four weeks reduced blood sugar levels by 49.16% (Ngatirah *et al*., 2020). Another study of diabetic patients proved consuming the yam for 14 days lowered their blood sugar levels (Sutriningsih and Ariani, 2017). Cheang *et al*. (2017) developed food products, i.e., noodles, from the yam that significantly reduced body weight, BMI (Body Mass Index), and abdominal circumference. Glucomannan, a water-soluble fiber, will form a gel when passing through the digestive tract and can be fermented easily in the large intestine. This process will trigger the production of glucagon-like peptide 1 (GLP-1) and peptide YY. GLP-1 is an incretin hormone that stimulates the pancreatic cells to secrete insulin (Susanti, 2014).

Roselle (*Hibiscus sabdariffa* L.), a species of the Malvaceae family, is widely used as food and medicine in Indonesia and an alternative therapy for patients with high blood pressure and cholesterol levels. Roselle also has numerous pharmacological activities: antibacterial, antifungal, anti-inflammatory, antidiabetic, antioxidant, hepatoprotective, and antihypertensive, thought to result from compounds such as polyphenols, alkaloids, tannins, flavonoids, saponins, organic acids, anthocyanins, and polysaccharides in all parts of the plant (Rocha *et al*., 2014). Its antioxidant character provides various therapeutical benefits, including an effective antidiabetic through several mechanisms of action. As confirmed by multiple studies (Wang *et al*., 2011; Kartinah *et al*., 2019), roselle lowers postprandial blood sugar, acts as a GLP-1 agonist, protects kidneys, and increases the regeneration of pancreatic beta cells (Oktaviani and Megantara, 2018).

There have been various studies of elephant foot yam. However, little is known about the potential utilization of this tuber crop by local communities in Indonesia. Besides, most of the yam products are exported to Japan. Innovations in developing yam-based products are undoubtedly needed to increase their uses at home. This is especially true given the high demand for well-accepted functional foods that can improve the quality of life of diabetic patients. Light and much-desired functional foods by people from various circles are in jelly form. This study was intended to produce high-fiber jelly powder from a mixture of elephant foot yam powder and roselle, which have a synergistic effect, as an alternative functional food for diabetic patients.

2. Materials and methods

2.1 Materials

The materials used in this research were elephant foot yam (*Amorphophallus oncophyllus*) flour (Wikonjac, Sidoarjo), roselle (*Hibiscus sabdariffa* L) powder (Material Medica in Batu, Malang), iota carrageenan (CV Nura Jaya, Surabaya), citric acid (Golden Sinar Sakti, Jakarta), sucralose (Kanbo, China), 96% ethanol (Merck, Germany), aquadest, essence and food colorant (Red Bell, Tangerang), Folin-Ciocalteu's phenol reagent (Merck, Germany), sodium carbonate ($NaCO_3$) (Merck, Germany), ABTS (Sigma, Singapore), phenol (Merck, Germany), sodium hydroxide (NaOH) (Merck, Germany), sodium sulfite ($NaHSO_3$) (Merck, Germany), potassium sodium tartrate tetrahydrate ($C_4H_{12}KNaO_{10}$) (Merck, Germany), 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid (ABTS) (Sigma, Singapore), 3,5-dinitrosalicylic acid (DNS) (Sigma, Singapore), and formic acid (Merck, Germany). The research equipment included glass beakers (Pyrex, Germany), containers, grinder/blender (Philips, Indonesia), mortar, centrifuge (Hettich Rotovix 32, Germany), stirrer (Thermo, Indonesia), analytical balance, filter paper, 40-mesh sieve, UV-VIS spectrophotometer (Shimadzu UV-1900), micropipette (Socorex, Switzerland), IR spectrophotometer (Agilent 630), and texture analyzer (Agrosta Texturometer Version 2, France).

2.2 Glucomannan extraction

Glucomannan was extracted from the elephant foot yam powder by leaching using ethanol with different grades of concentration, as shown in Figure 1.

2.3 Identification and qualitative measurement of glucomannan contents

The glucomannan extract was qualitatively analyzed using the Fourier transform infrared (FTIR) spectrophotometry (Agilent 630) by reading the graph within the range of 650-4000 cm^{-1} (Saputro *et al*., 2014) and colorimetry with glucose standards.

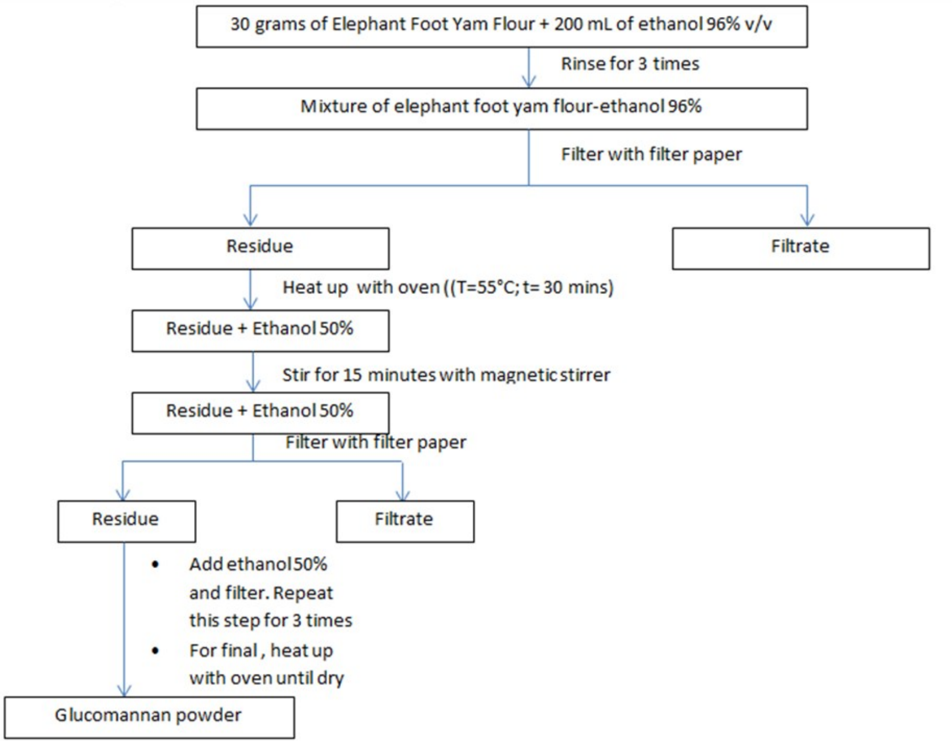

Figure 1. Elephant foot yam extraction flowchart.

2.3.1 Preparation of the reagent solution 3,5-dinitrosalicylic acid

Solution A was prepared by mixing 0.175 g of phenol with 375 mL of 10% NaOH and 175 mL of $NaHSO_3$. Solution B was made from 5.625 g of $C_4H_{12}KNaO_{10}$, 7.5 mL of 10% NaOH, and 22 mL of 1% DNS. Solutions A and B were then mixed and homogenized before being stored in a brown bottle to block light (Wardani *et al*., 2021).

2.3.2 Buffer preparation

A buffer was created from formic acid and NaOH. First, a total of 0.4 mL of formic acid and 24 mL of aquadest were poured into a 100 mL volumetric flask. Then, 20 mL of 0.5% NaOH was added to the flask (Wardani *et al*., 2021).

2.3.3 Preparation of standard glucose solution

Glucose weighing 0.25 g was dissolved in 25 mL of distilled water. The standard glucose solutions of different volumes (0.4; 0.30; 0.32; 0.10; 0.8 mL) and 0.8 mL of distilled water (as a placebo) were each put into a 10 mL volumetric flask and added with 0.6 mL of 1% DNS solution then stirred until homogeneous. This mixture was heated in a water bath at 100°C for 5 mins, stirred, and then added with distilled water to 10 mL. The absorbance was measured at a wavelength of 540 nm (Wardani *et al*., 2021).

2.4 Quantitative measurement of glucomannan content

2.4.1 Extract treatment

A sample of glucomannan extract weighing 0.2 g was dissolved in 50 mL of the buffer solution and then stirred at 30°C for 4 hrs. The buffer was added to the mix up to 100 mL and then stirred. Afterwards, the mixture was centrifuged at 4000 rpm for 20 mins to obtain the filtrate and supernatant (Wardani *et al*., 2021).

2.4.2 Preparation of glucomannan hydrolysate

Approximately 2 mL of glucomannan extract was poured into a 10 mL volumetric flask, added with 1 mL of 3M sulfuric acid, and homogenized. The mixture was later heated in a water bath for 1.5 hrs and then allowed to cool. Afterwards, it was added with 1 mL of 6M NaOH and distilled water to 10 mL, then homogenized (Wardani *et al*., 2021).

2.4.3 Analysis of glucomannan levels

Around 0.8 mL of glucomannan extract, hydrolysate, and aquadest (blank) were each put into a 10 mL volumetric flask, added with 0.6 mL of 3,5-dinitrosalicylic acid (DNS), and put in a water bath for 5 mins. Then, the solution was cooled to room temperature, then added with aquadest up to 10 mL. The absorbance value was measured at a wavelength of 540 nm. The glucomannan content was calculated using the formula below (Wardani *et al*., 2021):

$$\text{Glucomannan content (\%)} = \frac{5000f\,(5T0 - T)}{m}$$

where f is a correction factor (0.9), T is the weight (mg) of glucose in glucomannan hydrolysate, $T0$ is the weight (mg) of glucose in glucomannan extract, and m is the mass of the extracted glucomannan flour (200 mg).

2.5 Jelly formulation

In this study, three formulas of jelly were developed with different concentrations of elephant foot yam and roselle (Table 1). One gram of jelly powder was dissolved in 40 parts of water to make a jelly product. The scheme of the jelly-making process is depicted in Figure 2. Organoleptic (i.e., color, taste, aroma, texture) and physical parameters (i.e., gel strength and hardness), total phenolic contents, and antioxidant activities were measured to determine the best formula with the most favorable characteristics.

Table 1. Jelly formulation and composition.

Composition	Formula I	Formula II	Formula III
Glucomannan (g)	2.12	3.12	4.12
Carrageenan (g)	6.35	5.35	4.35
Rosella (g)	1.5	1.5	1.5
Citric acid (g)	0.03	0.03	0.03
Sucralose (g)	0.003	0.003	0.003

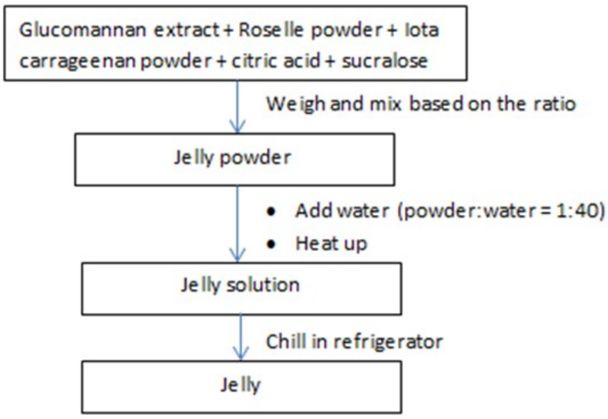

Figure 2. Jelly making flowchart.

2.6 Gel strength and hardness test

Gel strength was tested using a texture analyzer (Agrosta Texturometer Version 2). The measurements were repeated three times with three different points for each sample.

2.7 Measurement of total phenolic contents and antioxidant activity

2.7.1 Sample preparation

Approximately 1 g of the jelly produced was added with 20 mL of 96% ethanol and sonicated for 15 mins. The filtrate obtained was later filtered, put into a volumetric flask, and added with distilled water up to 25 mL.

2.7.2 Determination of total phenolic content

Total phenolic content was measured using the method explained in Almey *et al.* (2010) with modification. Gallic acid was used as a standard. One mL of the sample was added with 200 µL of the Folin-Ciocalteu reagent and 200 µL of 5% sodium carbonate (Na_2CO_3). The mixture was incubated for 15 mins at room temperature. Absorbance was measured using a spectrophotometer UV-Vis at a wavelength of 766 nm (Septiani *et al.*, 2017). The procedure was made in triplicates.

2.7.3 Antioxidant activity testing using the ABTS method

Before testing the antioxidant activity, an ABTS reagent was first prepared by dissolving 19.2 mg of ABTS powder and 3,500 mg of potassium persulfate with 5 mL of distilled water each. Then, both solutions were incubated in the dark for 12 hrs. The two solutions were mixed and added with distilled water up to 25 mL (Avanti *et al.*, 2021). Afterwards, for the antioxidant activity assay, 180 µL of the sample was put into a 96-well microplate and added with 20 µL of the ABTS reagent. The absorbance was measured using a microplate reader at a wavelength of 720 nm (Avanti *et al.*, 2021).

3. Results and discussion

3.1 Glucomannan extraction yield

The glucomannan extraction produced a 92.40% yield. In this study, glucomannan was purified from the elephant foot yam flour by leaching using ethanol with different grades of concentration: starting with 96% ethanol and then with 50% ethanol. The mixing process used a stirrer set at 300 rpm for 15 mins. This method aimed to remove impurities from the yam flour (Tatirat and Charoenrein, 2011). The residue obtained was dried in an oven at 60°C.

Elephant foot yam is a tuber plant that contains glucomannan, which should be derived through extraction prior to use. Glucomannan is a water-soluble hemicellulose (a type of polysaccharide) commonly sourced for functional foods with a high molecular weight of about 666.6 daltons (National Center for Biotechnology Information, 2023). One molecule contains D-mannose (67%) and D-glucose (33%). In water, 1% glucomannan creates a viscosity of 30,000 cP, which is categorically very high (Aryanti and Abidin, 2015). The viscous nature is associated with glucomannan's high water absorption: 1 g of glucomannan can absorb 100 g of water.

3.2 Qualitative presence of glucomannan in the extract

FTIR spectrophotometry was employed to ensure the presence of glucomannan in the derived extract using specific groups of glucomannan whose FTIR spectrum is generally found at a wavelength of 4000-400 cm^{-1}.

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Results showed several specific areas signifying the presence of glucomannan (Figure 3), which was the most visible in the range of 3000-3700 cm^{-1}, indicative of the OH group. This finding corresponds to Aryanti and Abidin (2015), who detected the OH group at 3400 cm^{-1}. In the current study, the C=O bond was shown at 1636.3 cm^{-1}, which is not too far from the frequency read by Darmawati *et al*. (2020), at 1653 cm^{-1}.

The CH_2 group commonly appears in the region flanked by 1375.4 and 1420.1 cm^{-1}. In Aryanti and Abidin (2015), the CH_2 group was found from the wavelength 1413 cm^{-1} to 1377 cm^{-1}. The C-OH bond was shown at 1149.9 cm^{-1} or very close to the one detected in Aryanti and Abidin (2015), at 1150 cm^{-1}. The 1,4-glycosidic bond of the cyclic ether (COC group) was seen at 1023.15 cm^{-1}, similar to the same bond in Darmawati *et al*. (2020): 1,4 glycosidic bond was detected at 1020.34 cm^{-1}.

Glucose-mannose markers in the glucomannan samples were observed in the region between 872.2 cm^{-1} and 943.01 cm^{-1}. According to Widjanarko *et al*. (2011), mannose and glucose markers are found within the range of 808.12 and 875.62 cm^{-1}, forming pyranose-characteristic of CH-stretch vibrations. Furthermore, some absorbance values showed a shift in wavenumbers and wave sharpness in the glucomannan functional group. Thus, it can be confirmed that the extract tested in the FTIR spectrophotometry contains glucomannan.

3.3 Glucomannan levels in the extract

Table 2 shows that the extract produced in this study contained 46.32% glucomannan. This result confirms that of Saputro *et al*. (2014), who reported that the levels of glucomannan after purification could vary from 36.69% to 64.22%. Glucose was used as a comparison reference in determining glucomannan concentrations because it is a monomer of glucomannan, which can thus provide accurate results. Figure 4 shows the absorbance values of glucose in various concentrations, from which the standard curve of the regression equation for glucose was obtained, y = 3.2114x - 0.0673. This regression equation was then used to calculate the levels of glucomannan in the extract and its hydrolysate. Both the glucomannan extract and hydrolysate were made in triplicate. Glucomannan was extracted using a stirrer at 300 rpm for four hours to remove insoluble substances such as starch and cellulose, increasing its overall solubility in water. To prepare hydrolysate, sulfuric acid was used to hydrolyze glucomannan into monomers: glucose and mannose. Adding DNS reagent and heating at the end of the reaction accelerates the reaction of glucose in the extract and hydrolysate to form color compounds for the spectrophotometry at 540 nm (Figure 5) (Rismawati *et al*., 2016).

Table 2. Glucomannan levels of the extract (in triplicate measurements).

No	Replication	% Glucomannan
1	I	44.99%
2	II	45.94%
3	III	48.04%
	Mean±SD	46.32±1.5607%

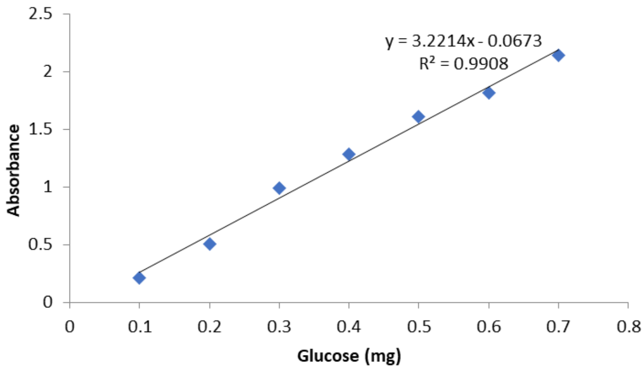

Figure 4. Standard curve of glucose.

3.4 Physical properties of the glucomannan jellies

Three formulas in Table 3 were developed to optimize the concentration and weight of glucomannan and iota carrageenan used in the jelly composition. These

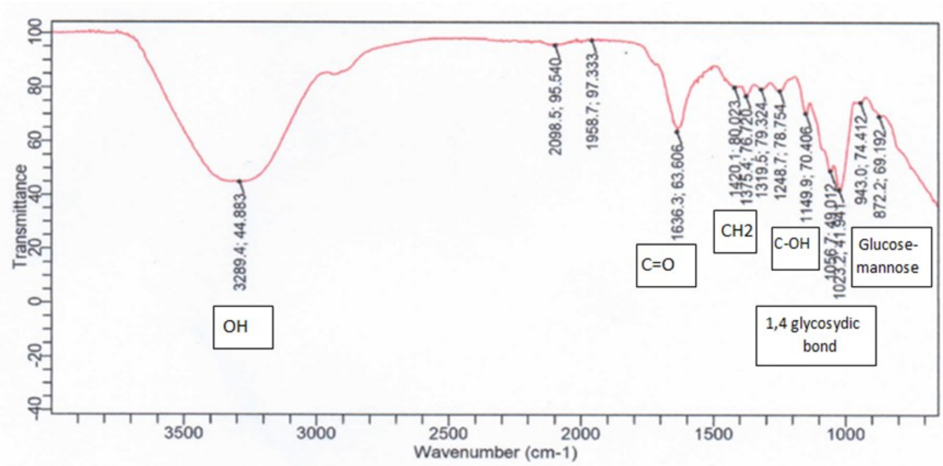

Figure 3. FTIR spectrum of glucomannan extract.

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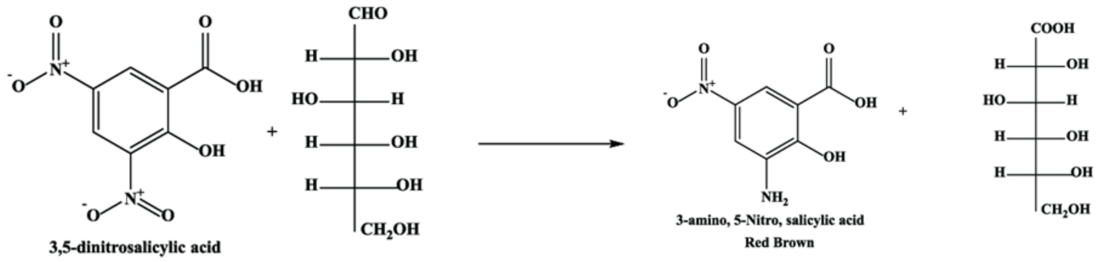

Figure 5. Reaction of 3,5 dinitrosalicylate and glucose. Source: Singh *et al*. (2021).

Table 3. Organoleptic properties of the jellies produced with three formulas.

No	Observation parameters	Formula I	Formula II	Formula III
1	Visual appearance			
2	Color	Brown-red	Brown-red	Brown-red
3	Taste	Sweet; slightly sour	Sweet; sourer than formulas I	Sweet; slightly sour
4	Odor	Not specific	Not specific	Not specific
5	Texture	Tender and firm; elastic or quivering when cut from the mold	Porridge-like but slightly firmer than formula III	Porridge-like but not sticky

formulas are different in the organoleptic specification for taste and texture. Iota carrageenan is a gelling agent used to produce an elastic soft-textured jelly that remains firm when chewed. It was added to the formula because increasing the concentration of glucomannan decreased the gel strength and, thus, hardness due to its high water-absorbing capacity.

3.5 Gel strength and hardness

Gel strength and hardness were tested to ensure that the jelly could be consumed with a spoon. Based on the results presented in Table 4, formula I produced a gel strength of 21.21 g/cm^2 and hardness of 24 cN (equivalent to 24 g), or the highest values among the formulas tested, which are believed to result from its smallest glucomannan and largest carrageenan content. The gel strength shares a similar characteristic to the one tested by Sinurat *et al*. (2006), 15±1.4 g/cm^2, which showed a firm gum that would hold its shape. A previous study by Kaya *et al*. (2015) on the glucomannan-carrageenan gel showed a hardness of 291.77 to 701.71 g. It is believed that the sharp difference between the current and results by Kaya *et al*. (2015) due to the latter used kappa instead of iota carrageenan.

Among the three formulas, formula I contained the largest carrageenan, resulting in the best texture. Based on Kaya *et al*. (2015), adding more carrageenan to the composition will increase the gel hardness due to the formation of a double helix. This will produce aggregates in large quantities of very strong nets, creating small intermolecular space and pushing out the free water. In other words, adding carrageenan induces a synergistic effect that reduces syneresis. As a result, gels with a stronger bond and a harder texture are formed (Sinurat *et al*., 2006). On the contrary, increasing the glucomannan percentage in the formulation decreases hardness because the high water-absorbing capacity of glucomannan allows more free water to fill the intermolecular space and soften the gel. Gel strength depends on the stiffness of the chain and the amount, type, and position of sulfate, which can induce or prevent the formation of the double helix chain (Suryani *et al*., 2015).

Table 4. Gel strength and hardness of jellies produced from different formulations.

Formula	Gel strength (g/cm^2)	Gel hardness (cN)
Formula I	21.21	24
Formula II	17.67	20
Formula III	14.14	16

In addition, citric acid has been reported to increase the gel's ability to bind water, resulting in a firm gel with a soft texture (Fajarwati *et al*., 2017). Sucralose was the citric acid added in a very small amount to the formula to lower the pH and enhance the flavor and texture. Sucralose is a synthetic sugar with a very low calorific value and a minor effect on blood sugar levels. It is 600 times sweeter than sucrose and is widely used in various food products, such as jam, ice cream, candy, and jelly. The FDA has also approved the use of sucralose as a sweetener.

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3.6 Total phenolic content and antioxidant activity

Tables 5 and 6 show the total phenolic content and antioxidant activity tested using the ABTS assay. The total phenolic content was calculated using gallic acid as the standard curve, from which a regression equation was obtained, y = 0.0656x + 0.0399, with a correlation value of 0.9973. Compared with the other two formulas, formula I had the highest phenolic content of 0.30%. At the same sample concentration, the ABTS assay showed that formula I also had the best antioxidant activity that reduced 90.51% radicals. These results indicated a correlation between total phenolic content and radical scavenging activity (Kumar and Goel, 2019). Phenolic compounds scavenge radicals with various mechanisms (Pereira *et al*., 2009).

Table 5. Total phenolic contents of jellies produced from different formulations (n = 3).

No	Formula	Total phenolic content (%GAE)
1	I	0.30±0.0168%
2	II	0.13±0.0178%
3	III	0.27±0.0214%

Table 6. Antioxidant activities of jellies produced from different formulations using the ABTS assay at 4 g/100 mL (n = 4).

No	Formula	Antioxidant activity (% inhibition)
1	I	90.51%
2	II	73.49%
3	III	88.16%

Antioxidant activity is one benefit that increases physiological functions, as expected from various functional food products. Besides, natural antioxidants provide many benefits for people with diabetes as they can reduce the risk of complications due to diabetes mellitus (Bajaj and Khan, 2012). Phenolic content and antioxidant activity analyses of the tree formulas determined whether or not the jellies produced could be further developed as a functional food for diabetic patients. It was found that all these formulas exhibited antioxidant activity, which is presumably due to the presence of the yam's glucomannan and roselle. Various clinical studies stated that glucomannan reduces random and 2-hour postprandial blood sugar levels after regular consumption for two to four weeks. Glucomannan can attract water in the digestive system, thereby increasing gel formation and viscosity, decreasing the rate at which food is absorbed in the small intestine, slowing gastric emptying, and inhibiting the absorption of cholesterol in the jejunum and bile acids in the ileum (Vuksan *et al*., 2000).

Roselle was added to the formulas to improve the jelly's taste and color. In addition to natural dye, roselle contains mainly anthocyanins and smaller concentrations of other groups of phenolic compounds. Many scholars have documented its various pharmacological activities for diabetic patients and linked them to anthocyanins. For instance, in Subhaswaraj *et al*. (2017), the ethanolic extract of roselle has an IC_{50} of 184.88 g/mL. Therefore, combining glucomannan with roselle is expected to increase the jelly's benefits, especially in controlling blood sugar.

The jelly produced in this study has different characteristics from previous studies. It was thought to result from variations in the composition of glucomannan and carrageenan and the type of carrageenan. Overall, the formula proposed and tested in this study (i.e., formula I) produced jelly that has good characteristics and is rich in phytonutrients, meaning that it has the potential as a functional food for diabetes mellitus patients.

4. Conclusion

Three functional food formulas of varying glucomannan and carrageenan concentrations have been tested in this study. Formula I, consisting of 2.12 g of glucomannan, 6.35 g iota carrageenan, 1.5 g roselle powder, 0.03 g citric acid, and 0.003 g sucralose, produces jelly with the most favorable characteristics. Based on physical observation, the jelly shows the best color and texture compared to the one made with formulas II and III. Also, according to the gel strength and hardness analysis, it has the most elastic and firm texture. Most importantly, formula I produces the highest total phenolic content and antioxidative activity. In conclusion, formula I is the best jelly formula.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgments

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