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FUNCTIONAL SUGAR FROM MANGO (*Mangiferaindica*) RICH IN ANTIOXIDANTS AND POLYPHENOLS POTENTIAL FOR ANTIDIABETIC

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ABSTRACT

This study was conducted to produce crystalized sugar from mango and determine vitamin C content, quercetin and kaempferol in it. Mango sugar extraction was carried out on 3 samples with different amounts (mangoflesh:water) for 3 samples respectively S1 (900:100), S2 (850:150) and S3 (800:200). Samples were blended and crystallized using freeze driyer (Lyovapor™ L-200**)** for 60 hours. Vitamin C analysis from 3 samples of mango sugar using titration iodometric method (mg/100g).Quercetin and kaempferol analysis using High Performance Liquid Chromatography (HPLC) technique at a wavelength of 415nm. Obtained the amount of vitamin C in S1 was 140,53mg. S2 was 136,65mg. S3 was 115,80mg. The formulation of mangoes with water in the highest amount of mango sugar containing vitamin C is S1. Obtained the amount of quercetin and kaempferol: S1 was 51.79µg/g and 49.84µg/g; S2 was 50.96µg/g and 50.34µg/g; S3 was 51.93µg/g and 49.78µg/g. There was a significant difference ($P<0.05$) that determined vitamin C levels between sample formulations. Mango has a big potency to be developed in to crystalized sugar. Mango sugar may be a great substitute for cane sugar, since antioxidants and polyphenols have the ability to improve lipid profile, stabilize blood glucose fluctuation and can be potentially antidiabetic sugar.

Keywords
Mango sugar, polyphenols, antioxidant, kaempferol, Antidiabetic.

Introduction

Mango (*Mangiferaindica L)* is a tropical fruit with a sweet taste and has become a favorite fruit in the world. It even becomes one quite essential fruit cultivated in more than 100 countries across the globe, particularly in Asia. Mango has a good taste, attractive color, exotic aroma, rich in nutrients and high carotene content, ascorbate acid, and phenolic compound as well as well known as the king of fruit in East Asia (Tang et al, 2019). The mango variety dominantly spread in Indonesia is mango*gadung*. It specifically has a thicker pulp than any other mango varieties (Sarinah*et al*, 2017).

The biggest producer of mango fruit is Thailand with a total of 18,779,000 tons per year. Indonesia positioned the 5th rank of the largest mango producing country with 2,184,399 tons per year (Low, 2019). In accordance with this data, Indonesia has the potential to develop the use of mangoes for various processed products. Mangoes contain nutrients, namely per 100 g is 272 kJ of energy, 17.00 g carbohydrates, 14.80 g sugar, 1.80 g fiber, 0.27 g fat, 0.51 g protein, vitamin A equiv. 38 mg, β-carotene 445 mg, thiamin (vitamin B1) 0.058 mg, riboflavin (vitamin B2) 0.057 mg, niacin (vitamin B3) 0.584 mg, pantothenic acid (vitamin B5) 0.160 mg, vitamin B6 0.134 mg, folate (vitamin B9) 14 mg, vitamin C 27.70 mg, calcium 10 mg, iron 0.13 mg, magnesium 9 mg, phosphorus 11 mg, potassium 156 mg, and zinc 0.04 mg. Mango pulp is also high in food

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fiber, vitamin E, ß-carotene, water, and polyphenols (Babarinde et al, 2019). The polyphenols contained in mangoes are those of Quercetin and Kaempferol types (Coelho et al., 2019; Suleman et al., 2019; Nurkolis et al., 2020). A meta-analysis study shows that the polyphenols of the Quercetin and Kaempferol types can improve Lipid profiles and stabilize blood glucose fluctuations (Bule et al., 2019; Dabeek et al, 2019; Tabrizi et al, 2019).

Several types of sugar are spread in society today, such as sugar (cane sugar) and brown sugar or palm sugar. A study shows that excessive and continuous sugar consumption that exceeds the recommended limit suggested by the Ministry of Health can significantly increase the risk of metabolic syndromes such as diabetes, coronary heart disease, obesity, and others (Fadhilah, 2012). Sugar (cane sugar) and brown sugar also do not contain polyphenols (Damayanthy, 2018; TKPI Update Team, 2018).

The World Health Organization (WHO) estimates that by 2030 there will be around 21.3 million Indonesians suffering from diabetes. According to the Indonesia Diabetes Association (Persadia), type 2 diabetes is the most common type of diabetes, which is about 95% of all DM cases. Type 2 diabetes is diabetes that is not related to insulin. Risk factors that influence the incidence of Type 2 diabetes include food intake or consumption such as the habit of frequently consuming sugar-sweetened drinks or foods such as soft drinks and fruit drinks (fruit drinks in packs) and less consumption of high levels of vegetables and fruits antioxidants and polyphenols (Fadhilah, 2012).

Seeing the potential that mangoes can be developed into various preparations with high potential health benefits, the present study was aimed to process the mango *gadung* into crystal sugar and determine the ash content, moisture content, quercetin, and kaempferol or polyphenols contained in it. This is useful to help find solutions in handling diabetes and other non-communicable diseases in Indonesia by looking at the polyphenols and antioxidants in this mango sugar product.

METHODS

Sample Preparation or Sugar Making (Crystallization

In this sample preparation, the type of mango *gadung* was chosen because it is the dominant type in Indonesia and has thicker pulp than other mango varieties. The Gadung Mango was cleaned and only the pulp of 2550g was taken. There were three sample variations with additional differences in moisture content. Sample one (S1) was 900g mango pulp plus 100 mL water. Sample two (S2) was 850g mango pulp plus 150 mL water. Sample Three (S3) was 800g Mango pulp plus 200 mL water. The water used as the addition had room temperature. The three samples (S1, S2, and S3) each of which was blended (Turbo, speed 2) for 10 minutes. The purpose of this variation was to determine whether there is an effect of adding water on crystallization results, the content of vitamin C and polyphenols (Quercetin and Kaempferol). Furthermore, the three samples (S1, S2, and S3) were put in the freezer overnight (± 12 hours) at a temperature of -22^0C before crystallization. Crystallization used the freeze-drying method for 60 hours with a pressure of 0.100 mbar and the samples were frozen. This crystallization process uses a freeze dryer in the Animal Husbandry Product Technology Laboratory of IPB, Bogor.

Determination of Polyphenol (Quercetin and Kaempferol) Content

Determination of polyphenol content used HPLC and was conducted at the 7th Floor Instrument (Physics) Laboratory, Sahid University, Jakarta. Determination of absorption wavelength of Quercetin and Kaempferol was conducted at the maximum rate. A total of 10 mg of Quercetin and Kaempferol were put in a 50-mL shaped flask dissolve and dilute with chloroform to the mark. Then, a 2.5 mL pipette is put into a 10-mL flask, diluted with chloroform to the mark. Furthermore, the spectrum was made using a UV-Vis spectrophotometer at a wavelength of

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450nm. Selection of mobile phase and flow rate A total of 10 mg of Quercetin and Kaempferol were put into a 50-mL flask, dissolve and dilute with chloroform to the mark. Then, a 2.5 mL pipette was put into a 10 mL flask, diluted with chloroform to the mark. Inject an amount of 20 µl into the HPLC device using the mobile phase methanol-chloroform (94: 6); methanol-tetrahydrofuran-water (67: 27: 6); chloroform-tetrahydrofuran-water (67: 27: 6); acetonitrile-chloroform (92: 8); and chloroform-tetrahydrofuran-methanol (70: 25: 5) with flow rates of 0.5 mL / minute and 1 mL / minute. The selected mobile phase and flow were those that provide the best separation with the least retention time.

Determination of Vitamin C Content

Determination of vitamin c levels used is by using the iodometric titration method and using the formula to calculate vitamin c levels:

$$\boldsymbol{vitamin\ c\ level\ (mg/100g) = \frac{V\ I2\ x\ 0,88\ x\ Fp\ x\ 100}{Ws\ gram}}$$

Determination of Water Content

The determination of water content used is the AOAC drying method (Thermogravimetry). The principle of this method is based on the evaporation of water in the material by heating, then weighing it to a constant weight. The weight reduction that occurs is the water content contained in the material. The way this method works was that the empty plate was heated in an oven at 1050 C for 30 minutes, cooled in a desiccator for 15 minutes, then weighed (W0). Then a 2-gram sample was put in a plate with known weight, weighed (W1), then dried in an oven at 1050 C for 3 hours, cooled in a desiccator for 15-30 minutes, then the plates and contents were weighed and dried again for 1 hour. , and cooled in the exicator, weighed again (W2). the water content was calculated using the following formula:

$$\boldsymbol{water\ content}(\%) = \frac{(\boldsymbol{W1 - W2})}{(\boldsymbol{W1 - W0})}\boldsymbol{x\ 100}$$

Determination of Ash Content

The procedure for determining the ash content was carried out using the AOAC 2005 method. Meanwhile, the procedure is as follows: the cup was dried in an oven at 105° C for 1 hour. Then, it was cooled in the cup for 15 minutes in a desiccator and weighed. 2 grams of the sample was put into a furnace where the temperature was 550° C for 3 hours. It was then cooled outside the furnace to a temperature of ± 120° C and put in a desiccator. The plates and ashes were weighed so that a constant weight was obtained. Calculation of the ash content was carried out using the following formula:

$$\boldsymbol{ash\ content}(\%) = \frac{\boldsymbol{wieght\ of\ bowl\ after\ heated - constant\ weight\ of\ empty\ bowl}}{\boldsymbol{sample\ weight}}\boldsymbol{x\ 100}$$

RESULT AND DISCUSSIONS

Result of Crystallization

Table 1. Result of Crystallization

Sample	Crystallization Treatment	Result (Gram)
S1	*Freeze Drying* 60 Jam	400 grams
S2	*Freeze Drying* 60 Jam	400 grams
S3	*Freeze Drying* 60 Jam	400 grams

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The Freeze Dryer method was chosen as the crystallization method (See Figures 1 and 2) for mangoes into granulated sugar because this method dries or crystallizes in frozen or cold conditions so that it can maintain the content of polyphenols and other bioactive compounds such as antioxidants in mangoes which are volatile (Bubba et al, 2019). In table 1, The three samples (S1, S2, and S3) had no difference in the crystallization results, which yielded 400 grams of mango sugar. The results of this test indicate that the addition of a multiple of water of 50 ml has no effect on the granulated sugar obtained. This is in contrast to that of Rumayar et al (2011) regarding the crystallization of sucrose in the manufacture of crystal sugar from palm sap, that there are differences in crystallization results for each addition of water, possibly due to differences in sample types and crystallization methods used.

Figure 1. *Freeze Drying* Initial Sample

Figure 2.*60th Hour Freeze Drying* Sample

Test Result of HPLC

Table 2.Kaempferol and Quercetin Content

Sample	Kaempferol (µg/g)	Quercetin (µg/g)
S1	49.84	51.79
S2	50.34	50.96
S3	49.78	51.93
Mean	49.98 ± 0.30	51.56 ± 0.52

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The three samples (S1, S2, and S3) each of which had 400 grams of mango sugar (in table 1), were tested for the quercetin and kaempferol content in them using HPLC (High Performance Liquid Chromatography) with a wavelength of 450nm. Table 2 shows, Thequercetin content in S1 yielded 51.79 µg / g, S2 50.96 µg / g and S3 51.93 µg / g. The average quercetin content of the three samples was 51.56 ± 0.52µg / g. While the content of kaempferol was at S1 49.84 µg / g, S2 50.34 µg / g, and S3 49.7 µg / g. It was found that the average kaempferol content was 49.9 ± 0.30 µg / g. It can be said that of the three types of samples (S1, S2, and S3), there is no difference in terms of yield or amount of mango sugar produced (Table 1). However, in the results of the polyphenol content test, there was a difference in their quercetin and kaempferol content. So, the addition or formulation of water content to the sample in multiples of 50 ml can affect the polyphenol levels in the resulting mango sugar (Table 2). It is hoped that further research will increase the variation in the water content of the sample by a multiple of 50 ml, in order to obtain a sample formulation that is high in polyphenols.

Comparison between mango sugar and cane sugar, brown sugar (Palm)

Table 3. Comparison between mango sugar and cane sugar, brown sugar (Palm)

Brown Sugar (Palm)	**Cane Sugar**	**Mango Sugar**
Quercetin: 00.00 ± 0.00 µg/g	**Quercetin: 00.00 ± 0.00 µg/g**	**Quercetin: 51.56 ± 0.52 µg/g**
Kaempferol: 00.00 ± 0.00 µg/g	**Kaempferol: 00.00 ± 0.00 µg/g**	**Kaempferol: 49.98 ± 0.30 µg/g**
Glycemic Index: ± 70	**Glycemic Index: ± 65**	**Glycemic Index : ± 51 (mango flesh)**

In accordance with the previous study, brown sugar or palm sugar does not contain polyphenols in the types of quercetin and kaempferol, as well as sugar cane (Damayanthy, 2018; TKPI Update Team, 2018) when compared to mango sugar which contains polyphenols of quercetin and kaempferol types, this is shown in table 3. Based on meta-analysis studies, quercetin and kaempferol polyphenols can increase HDL levels and reduce LDL levels, triglycerides, total cholesterol, and reduce blood sugar fluctuations (Bule et al, 2019). Another meta-analysis states that the polyphenols of the quercetin and kaempferol types can significantly reduce the incidence of obesity and other metabolic syndromes (Tabrizi et al, 2019).

Further, in table 3, the comparison of the glycemic index (GI) between brown sugar (palm sugar) and sugar cane with mangoes obtained a low GI value, namely in that of mangoes (Permana, 2015). However, a test to determine the GI value of mango sugar is needed so that the results of this comparison are more accurate. A meta-analysis of foods with low GI values proved effective in reducing glycatedhemoglobin (HbA1c), fasting glucose, BMI, total cholesterol, and LDL (Zafar et al, 2019). Based on the fact that this mango sugar contains polyphenols of the types of quercetin and kaempferol and the lower glycemic index value of mangoes compared to sugar cane and brown sugar (palm), this mango sugar has the potential to become an alternative sugar that can increase lipid profiles and stabilize blood glucose fluctuations.

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Test Result of Vitamin C Content in Mango Sugar

Table 4. Vitamin C Content in Mango Sugar

Sample	Vitamin C Content (mg/100g)
S1	140.53
S2	136.65
S3	115.80
Mean	130.99

In table 4, the average level of vitamin C produced is 130.99 mg / 100g, these results indicate that the level or level of vitamin C is deemed high by looking at the comparison from SNI 01- 3842-1995, the level of vitamin C for food is 50 mg. The results of the Annova statistical test showed that there was a significant difference between the vitamin c levels of the three samples with p <0.05, which means that the multiplication of 50 mL water addition not only affects the polyphenol content but also the vitamin c content. This high vitamin C also indicates that mango sugar is a source of antioxidants that are beneficial to health. One of the functions of antioxidants is as an antidiabetic (Marwati et al., 2020).

Test Result of Water content and Ash Content in Mango Sugar

Table 5. Water Content and Ash Content in Mango Sugar

Sample	(%) Ash Content	(%) Water Content
S1	1.95	1.95
S2	1.88	2.00
S3	1.70	2.09
Mean	1.84	2.01

The previous study has shown that the higher the ash content, the lower the quality of the sugar product, and the ash content indicates an inorganic material that will affect the color and hygroscopic properties of sugar (Swasitini et al, 2017). The limit of ash content in crystal sugar (palm sugar) according to SNI 01-2891-1992 is 2% (Swasitini et al, 2017). According to SNI 01-2891-1992, this mango sugar meets the quality requirements and proves that mango sugar has good quality. Because, in table 5 shows the ash content in mango sugar is 1.84% or less than 2%.
The Indonesian National Standard (SNI 01-6237-2000) for the water content contained in brown sugar cane is a maximum of 8% (National Standardization Body 2000). Cane brown sugar with a water content of more than 8% has a shelf life of 3-4 weeks, if stored for longer it will cause the sugar to melt and have a soft texture (Garusti et al, 2019). In SNI for mango sugar, there is no limit on the water content of crystal sugar from mangoes. According to SNI 01-6237-2000, mango sugar that meets SNI 01-6237-2000 has a longer shelf life than cane brown sugar which is only 3-4 weeks old. Because, table 5 shows that mango sugar has a water content of 2.01%.

CONCLUSION

The addition of water in multiples of 50 ml to the Freeze Drying of mango sugar has no effect on the number of mango sugar crystals produced. However, it affects the content of polyphenols quercetin and kaempferol. There was a significant difference (P<0.05) that determined vitamin C levels between sample formulations. Mango sugar is a sugar that has polyphenols with kaempferol content of 49.9 ± 0.30 µg / g and quercetin content of 51.56 ± 0.52µg / g, compared to sugar cane and brown sugar which do not have polyphenols. Based on the results of the water content and ash content tests, this mango sugar has good quality and has a shelf life of more than

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4 weeks. Therefore, mango has great potential to be developed into crystallized sugar. The content of polyphenols (Quercetin and Kaempferol) in mango sugar can be an alternative to sugar cane because polyphenols have the ability to improve lipid profiles and stabilize blood glucose fluctuations.

Suggestion

It is suggested for further research to modify this mango sugar by; Neutralizing the distinctive taste of the mango, Testing the levels of polyphenols (quercetin and kaempferol) in mangoes and comparing them with this mango sugar, Determining the Glycemic Index of this mango sugar, Increasing the variety of water composition: mango and determine the composition with the highest polyphenol content.

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Conflict of Interests

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