Analysis of vitamin C in probiotic yoghurt with the addition of podang urang mango (*Mangifera indica* L.)

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Abstract. Yoghurt is a functional drink that is beneficial to health. However, nutritional losses, e.g. vitamin C, occurs during the thermal process. Therefore, vitamin C supplementation is needed to replace the lost amount during processing by adding the extract of podang urang mango (*Mangifera indica* L.). The yoghurt was made by adding 3% (v/v) of starter culture (*Lactobacillus delbrueckii* subsp bulgaricus RRAM-01 and *Streptococcus salivarus* subsp thermophilus RRAM-01) and mango extract at 0%, 2%, 4%, 6%, 8% and 10% (v/v) in four replications. The determination of vitamin C content was performed using titration method. The results showed that heating process at 85-90 °C in 35 minutes lowered the amount of vitamin C in the final product. However, the addition of podang urang mango extract at 6%, 8% and 10% could increase the amount of vitamin C.

1 Introduction

Functional food become a very important issue and people awareness increase in consuming a good functional food which is supporting for their health [1]. Yoghurt drink is considered as one of functional food and can be used to improve and maximize digestive system of human body. Yoghurt is the most popular fermented dairy product all over the world [2]. Yoghurt obtained from fermented milk using *Lactobacillus bulgaricus* and *Streptococcus thermophilus* bacteria or other lactic acid bacteria as appropriate, with or without the addition of other foodstuffs and permitted food additives [3].

Some researchers have been conducted on yoghurt drink related to its processing or making technique [4]. Long processing of yoghurt will result in reduced physicochemical content of the yoghurt [5]. Sterilization causes loss of a number of vitamins, folic acid and causes denaturation of serum proteins. Long and unprotected yoghurt storage will result in

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reduced. Therefore, there needs to be supplementation in yoghurt to increase the content of vitamins lost during the manufacturing process.

Supplementation or innovation of yoghurt ingredient is important with the purpose for providing a variety nutritional and flavored value in yoghurt drink to make it more attractive to consumers, both children or adult to consume yoghurt. Today, consumers demanding food products with natural preservatives of high quality and good shelf-life stability. Natural preservatives may be more effective and efficient in protecting food products from deterioration by physical, chemicals and spoilage by microorganisms compared to synthetic additives [6]. Innovation of yoghurt drink can be obtained by utilizing ingredient that has high nutrition and not yet widely used in the surrounding environment, one of which is podang urang mango.

Podang urang mango is one of the local fruits that are abundant in Kediri. According to BPS-Statistics of Kediri Municipality [7], the mango production in Kediri was 647.30 tons in 2020. The highest fruit production occurs in the harvest season, which is in September until December [8]. Podang urang mango has a reddish yellow fruit skin color. Fruit with length of 12.5 cm and a width of 7 cm, the thickness of the flesh of the fruit is 2-2.5 cm. The color of the fruit meat is orange, soft textured, and the taste of the fruit is sweet [9]. Experts believe mangoes are a great source of carotenoids called beta cryptoxanthin, which is a good cancer-suppression ingredient. Mangoes are also rich of vitamins such as vitamins C and E and antioxidants [10].

High levels of vitamin C in podang urang mango is expected to be able to benefits for humans. Recently vitamin C is required for the prevention of scurvy and maintenance of healthy skin, gums, blood vessels, etc. [11]. Because of benefits, it is necessary to do research on the addition of extracts, podang urang extract on probiotic yoghurt to see the vitamin C value.

2 Materials and methods

2.1 Preparation of samples and extraction of podang urang mango [12]

Podang urang mango fruit obtained from the district of Banyakan Kediri. The making of podang urang mango extract is done by maceration using mixture of hexane:acetone:ethanol (2:1:1 v/v). Filtrate (macerate) is then added with 10.0 ml aquadest and extracted using a split funnel until 2 layers are formed. The polar layer is extracted again until a clear filtrate is obtained. The obtained non-polar layer is collected and evaporated until dry extract is obtained.

2.2 Preparation starter cultures [13]

Starter cultures that used in this research were *Lactobacillus delbrueckii subsp bulgaricus RRAM-01* dan *Streptococcus salivarius subsp thermophilus RRAM-01*. Starter rejuvenation is done by inoculating yoghurt culture as much as 10% into sterilized milk first on the autoclave with a temperature of 115°C for three minutes. Furthermore, it is incubated at a temperature of 37 °C for 18 hours until coagulation is formed so that a work culture is obtained.
2.3 Making probiotic yoghurt with the addition of podang urang mango extract [14]

Full milk is heated at 85-90 °C for 35 minutes, then cooled until the temperature reaches 40-45 °C. Lactobacillus delbrueckii subsp bulgaricus RRAM-01 and Streptococcus salivarius subsp thermophilus RRAM-01 are added to milk. Population of bacteria used more than 107 cfu mL⁻¹, incubated at 37 °C for 18-24 hours until produce coagulation or thickened (plain yoghurt). After that added podang urang mango extract as much as 0% (P1), 2% (P2), 4% (P3), 6% (P4), 8% (P5) and 10% (P6). Yoghurt is stored at room temperature (29-30 °C).

2.5 Analysis of vitamin C [15]

Provide 10 gram of sample (mango, milk or yoghurt) of each treatment on 100 mL of water. Add 5 drops amyllum indicator, then titration with iodine (I2) 0,01 N to blue. Vitamin C content can be calculated by formula:

\[
\text{Vitamin C} \left(\frac{mg}{100g}\right) = \frac{\text{Vol I2} \times 0.88 \times Fp \times 100}{W \text{ sample (g)}}
\]

Vol I2 : Volume Iodine (mL)  
0.88 : 0.88 mg Vit C  
Fp : Dilution Factors  
Ws : Sample Weight (g)

2.6 Statistical analysis

The experimental design used in addition of podang urang mango extract in yoghurt is completely randomized design (CRD) and the data was analyzed using Duncan’s post hoc test [16]. The vitamin C data were presented as mean with standard deviation from at least six independent experiments.

3 Results and discussion

Mango, especially podang urang mango plays an important role in balancing and improving the diet of human being by providing such of nutrition like energy, carbohydrates, sugar, dietary fiber, protein, vitamin A, vitamin C, calcium, magnesium, phosphorus, potassium [17]. Vitamins are group of organic complex compounds that the everybody needs in small quantities. Vitamins must be supplies from outside because the human body cannot synthesize them. One of important vitamin is Vitamin C [18].

Vitamin C plays a vital role in our lives because many reasons. First reasons, it contributes to the synthesis of the collagen tissue around bones, teeth, cartilage, skin, and damaged tissue. Second reasons, this vitamin is needed to activate various enzymes related to the nervous system, hormones, and detoxification of medicine and poison in the liver. Third reasons, its role as an antioxidant is well-known in society; its solubility enables it to work as antioxidant within our bodily fluids. Fourth reasons, Vitamin C increases the rate of absorption of iron, calcium, and folic acid. Fifth reasons, it reduces allergic reactions, boosts the immune system, stimulates the formation of bile in the gallbladder, and facilitates the excretion of various steroids [19].

Vitamin C is highly soluble in water and alcohol, and very easily to oxidized. The oxidation of Vitamin C occurs quickly in a base environment at especially high temperatures. Light and heat damage Vitamin C in fruits and vegetables [20]. Qualitative proof of vitamin
C can be done using the Iodometry method. Because this solution is able to reduce vitamin C. Vitamin C is a powerful and powerful reducing agent. Simple can be titrated with a raw solution of iodine as an oxidizer [21]. Thus, Then the content of vitamin C in figs and passion fruit can be determined by the method of Iodometri.

Vitamin C in podang urang mango extract 28.32 mg/100 g. Titratable acid in podang urang mango extract was 0.12 %. Its lower than those reported by Amiarsi and Mulyawanti [22] who found that vitamin C in mango was 33.62-58.59 mg/100 g and titratable acid was 0.11%. This acidity was caused by organic acid such as acetic acid, ascorbate, citrate, butyrate, and propionate [23].

Table 1. Vitamin C in milk

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Vitamin C (mg/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw Milk</td>
<td>0.77 ± 0.005</td>
</tr>
<tr>
<td>Pasteurized Milk</td>
<td>0.14 ± 0.006</td>
</tr>
</tbody>
</table>

From the Table 1 it was observed that Vitamin C in milk is 0.77 mg/100 gram. It slightly lower than National Dairy Council that describe vitamin C in milk was 0.94 mg/100 g. There was a decrease in vitamin C from raw milk to pasteurized milk. [24] reported that heat or sterilization damage the composition of vitamin C. The higher the temperature used for the sterilization process, the more vitamin C getting down. Saleh [25] also concluded that the most vitamin loss/ decrease in the process of processing milk with heat is vitamin C. milk processing with sterilization in polyethylene causing a decrease in vitamin C 50% greater than vitamin B12 loss and folic acid. Vitamin C also decreases if food is left warm or hot for too long. Vitamin C amount is significantly reduced if stored at room temperature [26].

Table 2. Vitamin C in yoghurt with the addition of podang urang mango extract

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Vitamin C (mg/100 g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>P1</td>
<td>0.22±0.05a</td>
</tr>
<tr>
<td>P2</td>
<td>0.25±0.06a</td>
</tr>
<tr>
<td>P3</td>
<td>0.25±0.06a</td>
</tr>
<tr>
<td>P4</td>
<td>0.35±0.06b</td>
</tr>
<tr>
<td>P5</td>
<td>0.35±0.06b</td>
</tr>
<tr>
<td>P6</td>
<td>0.38±0.05b</td>
</tr>
</tbody>
</table>

abDifferent letters in a column indicate statistically significant differences (P<0.05)

Table 2 showed that there is a decrease of vitamin C content in the yoghurt than in form of milk. Decreased vitamin C results in yoghurt influenced by several things, among others, in the process of making yoghurt, fermentation, and storage. In the process of making yoghurt occurs the process heating at so that it can damage vitamin C, while the way storage at less stable room temperatures affects damage Vitamin C [27].

Based on the measurement of vitamin C content, show that the increase of podang urang mango extract led to an increasing vitamin C content of this yoghurt. The addition podang urang mango extract at 6%, 8% and 10% significantly increasing vitamin content than 0%, 2% and 4%. This increase is due to an increase in the amount of podang urang mango extract.
that is added to yoghurt. The increase in the addition of podang urang mango also aims to improve the quality of organoleptic. Suitable with Suriasih [28] that increasing vitamin C levels in cow's milk yoghurt can also act as a flavor enhancer and can also be a special attraction for society (for consumers). Effendi [29] suggests that the main component in yoghurt is the acidic nature of lactic acid and its aroma substance lactic acid bacteria produced. However, with the addition of podang urang mango extract, aroma will also change.

Based on the data showed in Table 2, P2 (2%) dan P3 (4%) give not significantly different with P0 (yoghurt plain, without the addition of podang urang mango extract). This proves that vitamins are easily damaged components. After harvesting fruit then in the storage of a number of vitamins can disappear, depending on temperature, exposure to the air and sun, and the length of storage. The higher the temperature, the longer exposure to air and sun, the longer it is stored, the more missing vitamins [30].

There are not adequate amount of vitamins A and C in low fat dairy products, especially yoghurt drink which are not fortified. Usually, low fat milk and other dairy products (such as yoghurt drink) are enriched or fortified with vitamin A but not with vitamin C. Fortification of dairy products especially yoghurt drink with vitamins A and C leads to improvement in their nutrition quality and also increases their organoleptic quality [31]. Vitamin C stimulates the oxidation process and also stimulate the production of collagen which is essential for adrenal function. Vitamin C helps the body absorb micronutrients such as iron and calcium. This vitamin also helps in regeneration of vitamin E [32]. Vitamin C deficiency can lead to fatigue, immune body decrease, impaired osteoblast activation, reduced collagen synthesis in the body and triggering hair breakage.

4 Conclusion

The results showed that heating process at 85-90 °C in 35 minutes lowered the amount of vitamin C in the final product. However, the addition of podang urang mango extract at 6%, 8% and 10% could increase the amount of vitamin C.

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