Effect of Starch and Sulfuric Acid on Determination of Vitamin C in Papaya Fruit Using Iodimetri

Khoirul Ngibad¹, M. Sungging Pradana¹, Ingrid Retno Y. ¹

¹Department of Medical Laboratory Technology, Faculty of Health Science, Universitas Maarif Hasyim Latif, Sidoarjo, Indonesia

Abstract

Vitamin C is an antioxidant that can be used to inactivate oxidation reactions and prevent the formation of free radicals. Sources of vitamin C are fruits, such as papaya fruit. The purpose of this study was to determine the effect of the indicator volume of 1% starch and 2 N sulfuric acid on the determination of vitamin C in papaya fruit samples. This study used the iodimetri method with a standard iodine solution, starch indicator and the addition of sulfuric acid. The variations of starch indicator volume include: 0.25; 0.5, 1, 2 and 3 mL and the variations of sulfuric acid volume include: 0, 2, 4, 6, and 8 mL. The results showed that the optimum 1% starch indicator volume was 1 mL and the optimum volume of 2 N sulfuric acid was 2 mL.

Keywords

Vitamin C, sulfuric acid, starch, iodimetri.

INTRODUCTION

One of the vitamins needed by humans is vitamin C, also known as L-ascorbic acid. Vitamin C is a chemical compound with formula C₆H₈O₆. Vitamin C is an antioxidant, which is a compound that can act to inactivate oxidation reactions and prevent the formation of free radicals. On the other hand, vitamin C also can act as a calogen–forming compound (1).

Consumption of vitamin C that is not sufficient enough can cause the vitamin C deficiency. In contrast, excess of vitamin C intake in adolescents will also cause problems in the gastrointestinal system (2). Recommended Daily Allowance (RDA) is
the basis for adjusting the amount of adequate vitamins per day. The intake of at least 10 mg of vitamin C per day can prevent the occurrence of scurvy deficiency disease. According to the Regulation of the Minister of Health of the Republic of Indonesia No. 75 in 2013 concerning the Recommended Nutritional Adequacy Rate for the Indonesian Nation reveals that vitamin C is needed every day at least 40 to 50 mg (infants under 1 year), 40 mg (ages 1 – 3 years), 45 mg (ages 4 – 6 years), 45 to 50 mg (ages 7 – 12 years), 100 mg (pregnant women) and 150 mg (breastfeeding mothers) (3).

Sources of vitamin C are derived from fruits such as guava fruit, raisins, kiwi, lychees, oranges, persimmon, lime, papaya, strawberries, lemons, pineapples, melons, mangoes, star fruit, grapes, passion fruit, breadfruit, durian, avocado, jackfruit, pomegranate, banana, and watermelon. Determination of vitamin C in food and beverage packaging samples can be determined using the titrimetric method (4–7) and spectrophotometry (8–10). Among the two methods, the titrimetric method has the advantage of not requiring standard analyte solutions, simple equipment, easy to do and its accuracy is high.

Determination of vitamin C used the titrimetric method based on iodimerti, namely vitamin C will be oxidized by iodine standard solution and the endpoint of titration was indicated by the formation of blue color with the addition of starch indicators. The accuracy in determination of vitamin C was influenced by several measurement parameters, such as: volume of starch indicator and sulfuric acid used for the titration process. Some studies used variations of starch volume indicators: 2 drops (2), 3 drops (11), 2 mL (4), a few drop (3,12), and 3 mL (1). On the other hand, there are several studies that used the addition of sulfuric acid in the titration process (1,3,12). The uncertainty in the use of volumes of starch and sulfuric acid is feared to affect the accuracy in determining vitamin C levels in food and beverage ingredients. Thus, this study will know the effect of volume of the 1% starch indicator and 2 N sulfuric acid on the determination of vitamin C in samples of papaya fruit using iodimetri.

MATERIALS AND METHODS

Material

The materials used include Carica papaya L. (papaya fruit), 1% starch indicator, 0.01 N iodine standard solution, distilled water, filter paper, label paper, 0.01 N Na2S2O3, 0.01 N KIO3, 10% KI, and H2SO4.

Equipment

The equipments used include beaker glass, Erlenmeyer flask, volumetric flask, burette, statif, clamp, volume pipette, measuring pipette, scissors, knife, blender, spatula, and analytic balance.
**Procedure**

**Na$_2$S$_2$O$_3$ standard**

As much as 10 mL of 0.1 N KIO$_3$ solution was taken and then was put into the Erlenmeyer flask. Then, as much 5 mL of 10% KI solution and 2 mL of H$_2$SO$_4$ was added into the flask. Then, solution was titrated using the Na$_2$S$_2$O$_3$ solution until a light yellow color was formed. Next, mixture was added with 0.5 mL of 1% starch indicator and was titrated with Na$_2$S$_2$O$_3$ solution until a blue color disappears (12).

**I$_2$ standard**

As much as 10 mL of I$_2$ solution was taken into the Erlenmeyer flask. Furthermore, solutions was titrated using Na$_2$S$_2$O$_3$ solution to form a light yellow color. Next, mixture was added with 0.5 mL of 1% starch indicator. Then, mixtures was titrated with a Na$_2$S$_2$O$_3$ solution until a blue color disappears (12).

**Preparation of Sample**

Papaya fruit was shelled and the seed was removed. Then, it was cut into small pieces and was weighed as much as 100 grams. Furthermore, was blended until it resembles a slurry (juice). Then, mixtures was filtered with a filter cloth to separate between residue and filtrate. The residue was removed and the filtrate was put into 1000 mL volumetric flask and added with distilled water to the boundary mark. The filtrate obtained is ready to be measured using the iodimetric method.

**Effect of 1% starch indicator volume on the determination of vitamin C in papaya fruit**

As much as 25 mL of papaya sample solution was taken and put into Erlenmeyer flask. Then, solution was added by indicator solution of 1% starch with volume variation of 0.25; 0.5, 1, 2 and 3 mL. Then, mixtures was titrated with I$_2$ standard solution until a blue color was formed. From this treatment, the optimum volume of starch indicator will be obtained.

**Effect of addition of 2 N sulfuric acid on determination of vitamin C in papaya fruit**

As much as 25 mL of papaya sample solution were taken and put into Erlenmeyer flask. Then, as much as 1 mL of 1% starch indicator solution was added. Then, as much as 2 N H$_2$SO$_4$ solution were added with volume variations include 0, 2, 4, 6, and 8 mL. Then, mixtures were titrated with I$_2$ solution standard until a blue color was formed. From this treatment, the optimum volume of sulfuric acid will be obtained.

**RESULTS**

Effect of 1% starch Vitamin C levels in papaya fruit are tabulated in Table 1 and Fig.2. The obtained results revealed that papaya fruit contain maximum amount of vitamin C that is 12.91 mg/mL.
Table 1. Effect of 1% starch indicator volume on determination the vitamin C levels in papaya fruit

<table>
<thead>
<tr>
<th>Indicator volume (mL)</th>
<th>Titrant volume (mL)</th>
<th>Vitamin C levels (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n₁</td>
<td>n₂</td>
</tr>
<tr>
<td>0.25</td>
<td>1.1</td>
<td>0.9</td>
</tr>
<tr>
<td>0.5</td>
<td>0.6</td>
<td>1.1</td>
</tr>
<tr>
<td>1</td>
<td>1.4</td>
<td>1.5</td>
</tr>
<tr>
<td>2</td>
<td>0.9</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>1.4</td>
<td>1</td>
</tr>
</tbody>
</table>

Description: n₁ = test 1, n₂ = test 2, n₃ = test 3

Fig 1. Effect of 1% starch indicator volume on determination the vitamin C levels in papaya

In order to established methods for determination of vitamin C in papaya fruit, sulfuric acid was used in treatment (Fig.2 and Table 2). After incubation with sulfuric acid, a colored product is formed which is in a blue color.

Fig 2. Effect of sulfuric acid addition on the determination of vitamin C levels in papaya
Table 2. Effect of volume of 2 N sulfuric acid on determination of vitamin C levels in papaya

<table>
<thead>
<tr>
<th>Volume of sulfuric acid (mL)</th>
<th>Titrant volume (mL)</th>
<th>Levels of vitamin C (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n₁</td>
<td>n₂</td>
</tr>
<tr>
<td>0</td>
<td>0.8</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>0.8</td>
<td>0.9</td>
</tr>
<tr>
<td>4</td>
<td>0.8</td>
<td>0.6</td>
</tr>
<tr>
<td>6</td>
<td>0.8</td>
<td>0.8</td>
</tr>
<tr>
<td>8</td>
<td>0.8</td>
<td>0.8</td>
</tr>
</tbody>
</table>

Description: n₁ = test 1, n₂ = test 2, n₃ = test 3

DISCUSSION

The principle of determination the vitamin C contents using the iodimetric method is based on the reaction between vitamin C which is a strong reducing agent and standard solution of iodine which is an oxidizer with starch indicator. In this study, vitamin C in papaya fruit samples react with iodine standard solution so that an addition reaction occurs between iodine and double bonds of vitamin C on number 2 and 3 of carbon atoms become a single bond. If the whole vitamin C has been supplemented by standard iodine solution, then a reaction occurs between the droplets of the next iodine solution and the starch indicator solution so that iod–amilum was formed. The end point of the titration was indicated by the presence of blue color indicating that the titration has been completed because all vitamin C has been supplemented by iodine (5). Reactions that occur can be seen in Figure 3.

Fig 3. The reaction between vitamin C in papaya fruit with iodine produced the dehydroascorbic acid

Effect of 1% starch indicator volume on determination of vitamin C in papaya

In this study, the effect of 1% starch indicator volume on the determination of vitamin C levels in papaya fruit was studied using iodimetric titration. In the process of titration, the addition of an indicator serves to show that the endpoint of the titration has occurred so that the titration process must be stopped immediately. The addition of incorrect volume indicators is feared to affect the accuracy of the measurement of vitamin C levels in a sample.
Table 1 and Figure 1 showed that the addition of the indicator volume by 1 mL resulted the optimum vitamin C levels of 12.91 mg with the smallest standard deviation of 0.51. On the other hand, the addition of volume indicators of 1% starch as much as 2 and 3 mL decreased the vitamin C levels and increased the standard deviation value for addition the volume indicator of 1% starch by 3 mL. This is probably caused by the excess volume of 1% starch indicator, the less the volume of iodine standard solution needed so that the vitamin C levels in the sample reduced. Thus, the optimum volume of 1% starch indicator in determination the vitamin C in papaya fruit was 1 mL.

**The effect of addition of 2 N sulfuric acid on determination the vitamin C levels in papaya**

This study also was studied the effect of addition of 2 N sulfuric acid on determination of vitamin C levels in fruit Papaya using iodimetri. In the process of titration, the addition of sulfuric acid give an acidic condition in the solution of vitamin C when the titration process occurs with iodine standard solution. The effect of 2 N sulfuric acid on determination of vitamin C in papaya fruit was studied with 2 N sulfuric acid volume variations of 0, 2, 4, 6 and 8 mL.

Based on Table 2 and Figure 3, we can calculated that the addition of 2 mL of sulfuric acid volume produced the optimum vitamin C levels of 7.63 mg with standard deviation of 0.51. On the other hand, the more volume of 2 N sulfuric acid added caused slight decrease in fluctuating of vitamin C levels. In general, the standard deviation value in the study of the effect of sulfuric acid volume on determination of vitamin C levels in papaya fruit was smaller than the study of the effect of 1% starch indicator volume on the determination the vitamin C levels in papaya. It was also supported by visual observations in the laboratory proving that the color yield of the titration end point of all sulfuric acid addition treatments produced more stable color. This shows that at low pH vitamin C is more stable than high pH. Thus, the optimum volume of 2 N sulfuric acid on determination of vitamin C levels in papaya fruit was 2 mL.

**CONCLUSIONS**

The addition of 1% starch indicator and 2 N sulfuric acid volume had an effect on the determination of vitamin C levels in papaya fruit. The optimum 1% starch indicator volume obtained was 1 mL and the optimum volume of sulfuric acid 2 N was 2 mL.

**CONFLICT OF INTEREST**

No potential conflict of interest.
REFERENCES