Vitamin C – A Natural Antioxidant

Relation to topics / curriculum link:
• Nutrition in humans

Prior knowledge and skills needed:
• iodine test and DCPIP test
• meaning of redox reaction
• extraction of fruit juice
• simple titration method

Concept:

Vitamin C (ascorbic acid) is an antioxidant.

The amount of vitamin C content in food can be found out by titrating with an oxidising agent, e.g. iodine. In the reaction, vitamin C is oxidised, while iodine is reduced to iodide ions. The endpoint is determined by the formation of blue-black starch-iodine complex when all vitamin C is oxidised and excess iodine is free to react with the starch solution added as indicator.

Introduction

Vitamin C is an important component of a healthy diet. A well-balanced, healthy diet should ensure that there is enough vitamin C to prevent scurvy and other potential health problems. However, if taken in extreme excess, it may also cause harm to our body. Vitamin C is found in citrus fruits such as oranges, limes and grapefruits, and vegetables including tomatoes and green pepper. Vitamin C is easily damaged during food preparation. Moreover, the amount of vitamin C in most foods is usually high enough that the quantity remaining after processing is more than enough for a daily supply.

DCPIP is often used as the reagent to test for vitamin C. The amount of vitamin C in a food sample can be found out by titrating with DCPIP as vitamin C can reduce blue DCPIP to colourless. However, it is sometimes difficult to ascertain the endpoint of the titration due to incomplete decolourisation of DCPIP. In this experiment, iodine and starch solutions are used instead of DCPIP to find out the amount of vitamin C in food sample. When iodine solution is added to the food sample, vitamin C is oxidised, while the iodine is reduced to iodide ions. During the reaction, iodine added is immediately reduced to iodide as long as there is any vitamin C present. Once all the vitamin C has been oxidised, the excess iodine will react with the starch indicator, giving a blue-black
starch-iodine complex. This is the endpoint and it allows you to estimate the amount of vitamin C in the food sample.

**Materials**

- 0.1 % starch solution
- fruit / vegetable sample (e.g. green pepper, kiwifruit & orange)
- iodine solution (0.5 g iodine dissolved in 100 ml of 1% potassium iodide solution)
- vitamin C solution (1000 mg vitamin C tablet dissolved in 1 litre of distilled/deionised water)
- beaker
- dropper (1 ml)
- test tube
- test tube rack

**Activity: The amount of vitamin C in fruits and vegetables**

1. Label four test tubes for holding different sample solutions: vitamin C solution (C), green pepper (G), kiwifruit (K) and orange (O) juices. *(Please refer to p.4 for methods of extraction of fruit juices.)*
2. Add 1 ml of vitamin C solution and 5 drops of starch solution into the test tube labeled C.
3. Add iodine solution drop by drop into the test tube until the appearance of permanent trace of blue-black colour. Record the number of drops of iodine solution added. (Note: shake the test tube gently to mix the content after adding each drop of iodine solution).
4. Repeat steps 2 and 3 for three times and calculate the average number of drops of iodine solution added.
5. Calculate the amount of vitamin C in 1 ml of the vitamin C solution.
6. Repeat steps 2 to 4 for green pepper (G), kiwifruit (K) and orange (O) juices.
7. Tabulate the results and calculate the amount of vitamin C in 1 ml juice of each fruit / vegetable sample.
<table>
<thead>
<tr>
<th>Type of solution</th>
<th>Average number of drops of iodine solution added</th>
<th>Amount of vitamin C in 1 ml solution (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vitamin C solution</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Green pepper juice</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kiwifruit juice</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Orange juice</td>
<td></td>
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</tr>
</tbody>
</table>

**Questions for discussion:**

1. Would you get the same results if the experiment was repeated after several days using the same solutions? Why?
2. If the orange juice has been left in the refrigerator for a week, what will happen to the amount of vitamin C in the orange juice? Why?

**Further investigation**

1. **Investigating the amount of vitamin C in prepackaged fruit juices purchased from the supermarket**
   Some prepackaged fruit juices may contain less amount of vitamin C than that marked in the label. Design an investigation to find out if this is the case. Discuss the experimental design with your classmate before carrying out the investigation. Write a report of your investigation.

2. **Investigating the content of antioxidant in green tea**
   Most green tea manufacturers claimed that their products have rich antioxidant components, such as vitamin C. Design an investigation to compare the antioxidant content in different brands of green tea. Discuss the experimental design with your classmate before carrying out the investigation. Write a report of your investigation.

**Reference**

NOTES

Safety Precaution

Handle iodine solution carefully as it is an irritant.

Laboratory Preparation

I. Extraction of fruit juices

1. Method A

   Examples: orange, lemon

   • Cut the orange/lemon into two halves.
   • Squeeze one half of the orange/lemon by hand to force out the juice.
   • Filter the juice (to remove the solid tissue) using a filter funnel and a filter cloth. Collect the filtrate using a small beaker.

2. Method B

   Examples: kiwifruit, honeydew melon, grape

   • Remove the skin of the fruit.
   • Cut the fruit into small pieces on a white tile using a knife.
   • Grind the tissue by means of mortar and pestle to obtain about 10 ml of juice.
   • Filter the juice using a filter funnel and a filter cloth. Collect the filtrate using a small beaker.

II. Vitamin C is susceptible to oxidation by atmospheric oxygen over time. The sample solutions should be prepared immediately before the titration.

III. Identification of the endpoint in this titration is significantly affected by the colouration of the sample solution used. If the solutions are colourless or are pale in colour, there is no problem in identifying the endpoint. For juices with strong or deep colour, it may be difficult to identify the endpoint. It is advised to carry out a trial titration so as to acquaint yourself with the colour change at the endpoint. For some juices, it may just be darkening of the colour when arriving at the endpoint of the titration.