

Measuring vitamin C in fruit using a microscale titration

This resource is from the **Assessment for learning** series which can be viewed at: [rsc.li/44jTX18](https://www.rsc.li/44jTX18). The series contains lesson plans and associated resources to actively involve students in their learning.

Learning objectives

- 1 Safely carry out a simple microscale titration reaction.
- 2 Make inferences from experimental data and compare the amount of vitamin C levels present in different fruits.
- 3 Evaluate the results of the practical in terms of errors.
- 4 Plan an experiment to determine how vitamin C levels are affected by different conditions.

The practical allows learners to safely carry out microscale titration reactions and make inferences about the amount of vitamin C present in fruit and juices (LO1 and 2). Completion of the follow-up questions will allow learners to succeed in LO2 and 3. LO4 relates to planning an experiment as a group, which learners can carry out during a later session.

Note: learners may be unfamiliar with the word 'inferences' used in the learning objectives. Explain this as 'a conclusion reached using the evidence collected from a practical and your scientific knowledge'.

Introduction

Ascorbic acid, the compound we call vitamin C, dissolves in water. It is easy to take into our bodies via a solution of the compound, such as that found in fresh fruit juice. It is a key nutrient required in the body for wound healing and iron absorption. It also acts as an antioxidant to protect cells and helps support the immune system to fight infection. A deficiency in vitamin C can cause scurvy.

In this practical activity, learners work in groups to analyse different samples of fresh fruits and juices, using a simple microscale titration with iodine to determine the amount of vitamin C present. When ascorbic acid reacts with iodine, dehydroascorbic acid and hydrogen iodide are formed. Iodine is brown in colour, but the ions of iodide in the product are colourless.

By adding starch to the solution, the colour change observed is more distinct as the iodine and starch molecules combine to make a blue-black complex. This allows learners to determine when all the iodine has reacted with the ascorbic acid, as when all the iodine has reacted with ascorbic acid, there will be no blue-black complex left. This is called the endpoint of the reaction.

Scaffolding

There are two versions of the student worksheet: scaffolded (★) and unscaffolded (★★). The scaffolded sheet offers more support to allow learners to access the questions. Hints are found after some of the questions to support learners further and guide their answers.

Teaching sequence

Introduction

1. Invite suggestions about where vitamin C comes from, together with ideas about how much vitamin C is present in the source suggested.
2. Organise suggestions in a primitive table – 'lots', 'some' and 'little vitamin C'.
3. Introduce the practical, to test different fruits and juices for their vitamin C levels, and the learning objectives. Explain that the test will confirm or change some of the suggestions made.

Testing samples to determine vitamin C levels

1. Organise learners into groups of three or four, with four samples to test per group. Note: the same fruit or juice can be tested more than once by different groups, giving multiple results for reliability.
2. Demonstrate the technique for testing the samples.
3. Circulate and supervise the groups as they extract juice from fresh fruit and test the juice samples.
4. Collate the results.

Example results table

Fruit juice/vitamin tablet	Number of drops needed to react with all the iodine
100 mg vitamin C tablet in 100 cm ³ water	15
Lemon juice	30
Lime	35
Orange juice from one-day open carton	40
Kiwi	15
Melon	20
Mango	30

Method

A full method is given in the student worksheet, available to download here: rsc.li/3hieF5q.

Technician notes

Read our standard health and safety guidance (rsc.li/3zyJLkx) and carry out a risk assessment before running any live practical.

Equipment

Apparatus

- Safety glasses
- Droppers or plastic pipettes, one for each juice
- Test tubes, one per test, allowing six per group
- Test tube rack
- 10 cm x 10 cm white card for background
- Juice extractor and beakers for juice if fresh fruit is used

Chemicals

- Fruit juices to test, with packaging if the fruit is not fresh
- Water, about 100 cm³ per group
- A solution of 0.05 mol dm⁻³ iodine in potassium iodide
- Starch solution
- Vitamin C solution

Preparation

See CLEAPSS recipe sheet [RB050](#), refer to [SSERC](#) or contact your local safety advisory body for preparing iodine solutions.

- A solution of 0.05 mol dm⁻³ iodine in potassium iodide is prepared using:
 - Iodine, 1.27 g (HARMFUL, toxic to aquatic life)
 - Potassium iodide, 1.5 g
 - Distilled water, 100 cm³
- 0.1% starch solution
- 1 mg cm⁻³ vitamin C solution, prepared using:
 - Water, 100 cm³
 - Vitamin C tablet, 100 mg, to be dissolved in the water

Safety and hazards

- Wear safety glasses.
- Solid iodine (used in the preparation) is harmful in contact with the skin, harmful if inhaled and very toxic to aquatic life. See CLEAPSS Hazcard [HC054](#), refer to [SSERC](#) or contact your local safety advisory body.
- A solution of 0.05 mol dm^{-3} iodine in potassium iodide (used in the experiment) is not currently classified as hazardous.

Reviewing the results

When the groups have completed their tests, review their results. In a class discussion, review all the results and:

1. Use any duplicate results to help students understand about the reliability of test results.
2. Refer to juice packaging materials to give background information.

Investigating conditions affecting vitamin C levels

1. Introduce the next activity in which students will plan an investigation to explore how vitamin C levels are affected by different conditions.
2. Give each group a different investigation to plan, using the basic test as a starting point. Suggested investigations include:
 - How does temperature affect the amount of vitamin C?
 - How does light affect the amount of vitamin C?
 - How do packaging and storage conditions affect the amount of vitamin C?
 - How does a 'slow release' vitamin C tablet work?
 - Do fruits of different varieties (e.g. mandarin oranges, navel oranges, clementines, satsumas, etc.) contain different amounts of vitamin C?
 - Does frozen, concentrated orange juice contain as much vitamin C as fresh juice?
 - Which fruit contains the most vitamin C per 100 g? Make a thorough comparison of as wide a range of fruit varieties as possible.
3. Agree the criteria for peer assessment of investigation plans.
4. Give each group access to information sources and presentation materials.
5. Circulate and support as learners:
 - plan their investigations
 - prepare a presentation of their plan
 - elect a spokesperson to feedback to the class

Plenary

1. Hear each presentation.
2. Give feedback based on the quality of the proposed investigations and their scientific content.
3. Make sure that each learner makes a written record of the planned investigation.

Learners can carry out their investigations in a later session or at an after-school club.

Answers

1. Based on the experimental results... because it required the least drops of fruit juice to completely react with the iodine solution.
2. Based on the class results.
3. It must be added dropwise so that an accurate number of drops can be counted for when all the iodine has fully reacted.
4. Overshooting the end point, different sized drops, misjudging the colour change.
5. Based on juices used in the practical.
6. Based on juices used in the practical.
7. Results may differ as companies will have used quantitative analytical techniques, not qualitative. They will have used more accurate equipment.
8. Temperature, light, packaging and storage, different fruit variety, frozen vs. fresh, oxygen exposure, pH.

Extension questions

9. If 100 mg is in 100 cm³, then there is 1 mg in 1 cm³. To fully react with the iodine solution, 15 drops were added.
 - Lemon juice required 30 drops, therefore $15/30 = 0.5 \times 1 \text{ mg} = 0.5 \text{ mg}$
 - Lime required 35 drops, therefore $15/35 = 0.429 \times 1 \text{ mg} = 0.429 \text{ mg}$
 - Orange juice from one-day open carton required 40 drops therefore $15/40 = 0.375 \times 1 \text{ mg} = 0.375 \text{ mg}$
 - Kiwi required 15 drops, therefore 1 mg
 - Melon required 20 drops, therefore $15/20 = 0.75 \times 1 \text{ mg} = 0.75 \text{ mg}$
 - Mango required 30 drops therefore $15/30 = 0.5 \times 1 \text{ mg} = 0.5 \text{ mg}$

Alternatively, learners may use ratios and the inversely proportional relationship, e.g.

- Lemon juice required 30 drops, therefore $30/15 = 2$, inverse relationship gives $\frac{1}{2} = 0.5$, therefore $0.5 \times 1 \text{ mg} = 0.5 \text{ mg}$

Either method of calculation is acceptable.

10. All results above are for 1 cm³. Multiplying these by 100 would give the amount of vitamin C in 100 cm³

Fruit juice/vitamin tablet	Mass of vitamin C in mg in 1 cm ³	Mass of vitamin C in mg in 100 cm ³
Lemon juice	0.5	50
Lime	0.429	42.9
Orange juice from one-day open carton	0.375	37.5
Kiwi	1	100
Melon	0.75	75
Mango	0.5	50

Therefore, all fruit juices apart from orange juice from a one-day open carton would give you your daily recommended allowance of vitamin C.

11. Fruits from the table above with a high vitamin C content. Some consideration may be given to the lifespan of the fruit/fruit juice.
12. Answers may include some of the following points:
- The Admirals were right to consider a healthy diet to protect sailors against scurvy. Scurvy is caused by a deficiency in vitamin C, therefore it is an essential part of a healthy diet.
 - Fresh fruit contains the highest amount of vitamin C. Named fruits from either the experimental data or the answer to question 11 could be given, for example, kiwi and melon.
 - Fresh fruit would quickly spoil so is only a short-term solution during a sea voyage. It should be prioritised when visiting, or shortly after leaving, a port. Similarly with fruit juice.
 - Preserved fruit does contain vitamin C, although at a lower level to fresh fruit. Learners may refer to their answer to question 8, as the method of preservation (e.g. drying) will reduce the amount of vitamin C in the fruit.
 - Vitamin C is water soluble so can be lost through the process of cooking in water, if the cooking water is discarded.
 - Food storage should be carefully considered to reduce factors that affect the amount of vitamin C. Learners should refer to their answer to question 8.