Accuracy of Labeling Vitamin C in Orange Juice

Topic
Vitamin C can be extracted from commercially prepared orange juice to check the accuracy of labeling.

Introduction
Vitamin C, also known as ascorbic acid (see Figure 1), is an essential nutrient in the human diet. Vitamin C is used in several metabolic processes and is a cofactor for many important enzymes in the body. In addition, vitamin C is a powerful antioxidant that helps protect the body from disease. Minor deficiencies in vitamin C can cause a multitude of side effects, and major deficiencies can result in scurvy, a disease characterized by tiredness, joint and muscle pain, and bleeding gums. Before the disease was understood, sailors who were at sea for several months would develop scurvy unless lemons and limes were on board. Because vitamin C is water soluble, it is not made by or stored in the body. Excess vitamin C cannot be saved in the tissues and is simply excreted in the urine, so this nutrient must be consumed daily as part of a healthy diet. Good dietary sources of vitamin C include fruits, such as strawberries and mangoes, and green vegetables, such as broccoli, leafy greens, and Brussels sprouts.

![Chemical structure of ascorbic acid (vitamin C)](image)

Figure 1
Chemical structure of ascorbic acid (vitamin C)

Because of the importance of vitamin C to a balanced diet, some food manufacturers add ascorbic acid to their products. Even though oranges
contain a great deal of natural vitamin C, producers of orange juice commonly add ascorbic acid in order to raise the nutritional value. The amount of vitamin C in orange juice is reported on the nutrition label (see Figure 2). In this experiment, you will extract vitamin C from different brands of orange juice and compare the results to the amount listed on the nutrition label. You will use the technique of titration, a method of analysis used to find the exact quantity of a reactant in a titration flask. A burette is a calibrated tube with a stopcock that can be used to deliver a reactant to the flask (see Figure 3).

**Nutrition Facts**

<table>
<thead>
<tr>
<th>Serving Size 8 fl oz (240 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serving Per Container 8</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Amount Per Serving</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Calories</td>
<td>110</td>
</tr>
<tr>
<td>% Daily Value*</td>
<td></td>
</tr>
<tr>
<td>Total Fat</td>
<td>0g</td>
</tr>
<tr>
<td>Sodium</td>
<td>0mg</td>
</tr>
<tr>
<td>Potassium</td>
<td>450mg</td>
</tr>
<tr>
<td>Total Carbohydrate</td>
<td>26g</td>
</tr>
<tr>
<td>Sugars</td>
<td>22g</td>
</tr>
<tr>
<td>Protein</td>
<td>2g</td>
</tr>
<tr>
<td>Thiamin</td>
<td>10%</td>
</tr>
<tr>
<td>Folate</td>
<td>15%</td>
</tr>
<tr>
<td>Not a significant source of calories from fat, saturated fat, trans fat, cholesterol, dietary fiber. Vitamin A, calcium and iron. Percent Daily Values are based on a 2,000 calorie diet.</td>
<td></td>
</tr>
</tbody>
</table>

**Figure 2**

Sample nutrition label

**Time Required**

90 minutes

**Materials**

- 4 different brands of orange juice with no pulp
- four 400-milliliter (ml) beakers
- stirring rod
• four 100- to 250-ml flasks
• burette
• ring stand with burette clamp
• 100-ml graduated cylinder
• 1 percent starch solution
• dropper
• Lugol’s iodine solution
• labeling tape
• graph paper
• science notebook
Accuracy of Labeling Vitamin C in Orange Juice

Safety Note
Iodine will stain clothing and skin, so use extreme caution. Handle glassware carefully. Please review and follow the safety guidelines.

Procedure

1. Obtain samples of four different brands of orange juice.
2. Examine the label of each brand of juice to find the amount of vitamin C listed per serving. Record this information on Data Table 1.
3. Answer Analysis questions 1 and 2.
4. Measure 100 ml of orange juice sample A using a graduated cylinder. Pour the sample into an Erlenmeyer flask. Label the flask as “A”.
5. Add 10 drops of 1 percent starch solution into the flask.
6. Set up the burette on a ring stand in a burette clamp.
7. Fill the burette with Lugol’s iodine solution. Record the starting reading, which is the level of iodine in the burette, under Trial 1 on Data Table 2.
8. Titrate the orange juice slowly. To do so, open the stopcock to add one drop of Lugol’s iodine solution at a time, then swirl. Continue titrating until the solution in the flask turns a faint blue color that persists after swirling. When the blue color persists, you have reached the endpoint. (If the solution turns dark blue or purple, too much iodine has been added.) Record the level of iodine left in the burette, then calculate the amount of iodine used. Record the calculations on Data Table 2 under “Trial 1.”
9. Repeat steps 4 through 8, using a fresh sample from the same container until you have two good (light blue) titrations. (You may need to carry out several trials to get two good titrations.)
10. Repeat steps 4 through 9 with the other three samples of orange juice.
11. Average the amount of iodine used to titrate the two best trials from each sample. Record the averages on Data Table 2.
12. Answer Analysis questions 3 through 7.

Analysis

1. According to the labels, which brand of orange juice has the most vitamin C?
### Data Table 1

<table>
<thead>
<tr>
<th>Brand of orange juice</th>
<th>Amount of vitamin C (from label)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A.</td>
<td></td>
</tr>
<tr>
<td>B.</td>
<td></td>
</tr>
<tr>
<td>C.</td>
<td></td>
</tr>
<tr>
<td>D.</td>
<td></td>
</tr>
</tbody>
</table>

### Data Table 2

<table>
<thead>
<tr>
<th>Brand</th>
<th>Trial 1</th>
<th>Trial 2</th>
<th>Trial 3</th>
<th>Trial 4</th>
<th>Av. of two best trials</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Readings start/end</td>
<td>Am. of iodine</td>
<td>Readings start/end</td>
<td>Am. of iodine</td>
<td>Readings start/end</td>
</tr>
<tr>
<td>A</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
2. Using the data from Data Table 1, create a bar graph comparing the amount of vitamin C in each of the four samples as indicated on the labels.

3. Which type of orange juice required the most iodine in the titration, and therefore had the most vitamin C?

4. Using the data from the average column of Data Table 2, create a bar graph comparing the amount of vitamin C according to your experimental results.

5. How do your experimental results compare to the amount of vitamin C listed on the orange juice nutrition labels?

6. What could have caused the discrepancies between the actual amount of vitamin C from the experiment and the amount listed on the labels?

7. List some sources of error in this experiment that may have affected your results.

What’s Going On?

Titrations are performed to determine the unknown concentration of one substance using a substance with a known concentration. They are often done as neutralization reactions between an acid and a base. In this experiment, the ascorbic acid (vitamin C) in the orange juice reacted with the iodine that was titrated from the burette. As iodine reacted with ascorbic acid, the iodine molecules changed into colorless iodide ions. When all of the ascorbic acid had reacted with iodine, it began to react with the starch that was added to the orange juice, producing a blue-black starch-iodine complex. The slightly blue color of the solution at the titration endpoint indicated that all of the ascorbic acid had reacted. Therefore, the orange juice solution that required the most iodine in order to reach an endpoint had the highest quantity of vitamin C.

Ascorbic acid and iodine undergo an oxidation/reduction (or redox) reaction. This type of reaction always occurs in chemical pairs, with one chemical receiving electrons and another losing electrons. The molecule that loses electrons is oxidized, while the one that gains is reduced. During the reaction, the ascorbic acid lost electrons, which were transferred to the iodine molecule. The ascorbic acid was oxidized to dehydroascorbic acid while the iodine molecule gained electrons and was reduced to iodine ions.
Connections

Nutritional labels on food products in the United States must report the amount of certain nutrients, including vitamin C, in packaged food to the Food and Drug Administration (FDA). The FDA requires that nutrition labels on food packaging be accurate so the reported amounts of nutrients are tested frequently using methods much like the one used in this experiment. Most packaged foods are monitored by quality controllers directly after production. Nutrient values of most foods do not change very much from the time of production until purchase; therefore nutrition labels are very accurate. Vitamin C, however, is very reactive and breaks down easily with exposure to high temperatures, light, and water. Therefore, even if the amount of vitamin C is accurate during production, the actual amount that is in orange juice when it is consumed may vary depending on the shipping process and the length of time the juice is on the shelf at the supermarket.

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OUR FINDINGS

ACCURACY OF LABELING VITAMIN C IN ORANGE

Idea for class discussion: Find out how many students have a glass of orange juice every day. Ask them to suggest some of the health benefits of orange juice.

Notes to the teacher: To prepare a 1 percent starch solution, add 0.5 grams (g) soluble starch to 50 milliliters (ml) of distilled water heated to near-boiling. Mix well and cool before using.

Analysis

1. Answers will vary depending on the brands chosen.

2. Student graphs should include four types of orange juice along the X-axis and the vitamin C content along the Y-axis. The vitamin C content of each type of juice should be represented by a bar of appropriate height.

3. Answers will vary based on experimental results.

4. Student graphs should include the four types of orange juice along the X-axis and the amount of iodine used along the Y-axis. The amount of iodine used in the titration for each type of juice should be represented by a bar of appropriate height.

5. Answers will vary. Students should compare the amount of vitamin C listed for each brand of juice to the amount of iodine required in the titration.

6. Answers will vary. Vitamin C is very reactive and will break down easily during shipping and time on the shelf at the grocery store. The amount of vitamin C can be impacted by the exposure to light, high temperatures, or exposure to air.

7. Answers will vary. Some sources of error include: over-shooting the titration, which causes the solution to turn darker than it should be at the endpoint; improper reading of the iodine levels in the burette; and inaccuracies in the measurement of orange juice that was added to the flask before testing.
Each experiment includes special safety precautions that are relevant to that particular project. These do not include all the basic safety precautions that are necessary whenever you are working on a scientific experiment. For this reason, it is absolutely necessary that you read and remain mindful of the General Safety Precautions that follow. Experimental science can be dangerous, and good laboratory procedure always includes following basic safety rules. Things can happen very quickly while you are performing an experiment. Materials can spill, break, or even catch fire. There will be no time after the fact to protect yourself. Always prepare for unexpected dangers by following the basic safety guidelines during the entire experiment, whether or not something seems dangerous to you at a given moment.

We have been quite sparing in prescribing safety precautions for the individual experiments. For one reason, we want you to take very seriously every safety precaution that is printed in this book. If you see it written here, you can be sure that it is here because it is absolutely critical.

Read the safety precautions here and at the beginning of each experiment before performing each lab activity. It is difficult to remember a long set of general rules. By rereading these general precautions every time you set up an experiment, you will be reminding yourself that lab safety is critically important. In addition, use your good judgment and pay close attention when performing potentially dangerous procedures. Just because the book does not say “Be careful with hot liquids” or “Don’t cut yourself with a knife” does not mean that you can be careless when boiling water or using a knife to punch holes in plastic bottles. Notes in the text are special precautions to which you must pay special attention.

**GENERAL SAFETY PRECAUTIONS**

Accidents caused by carelessness, haste, insufficient knowledge, or taking an unnecessary risk can be avoided by practicing safety procedures and being alert while conducting experiments. Be sure to
check the individual experiments in this book for additional safety regulations and adult supervision requirements. If you will be working in a lab, do not work alone. When you are working off-site, keep in groups with a minimum of three students per groups, and follow school rules and state legal requirements for the number of supervisors required. Ask an adult supervisor with basic training in first aid to carry a small first-aid kit. Make sure everyone knows where this person will be during the experiment.

PREPARING

- Clear all surfaces before beginning experiments.
- Read the instructions before you start.
- Know the hazards of the experiments and anticipate dangers.

PROTECTING YOURSELF

- Follow the directions step by step.
- Do only one experiment at a time.
- Locate exits, fire blanket and extinguisher, master gas and electricity shut-offs, eyewash, and first-aid kit.
- Make sure there is adequate ventilation.
- Do not horseplay.
- Keep floor and workspace neat, clean, and dry.
- Clean up spills immediately.
- If glassware breaks, do not clean it up; ask for teacher assistance.
- Tie back long hair.
- Never eat, drink, or smoke in the laboratory or workspace.
- Do not eat or drink any substances tested unless expressly permitted to do so by a knowledgeable adult.

USING EQUIPMENT WITH CARE

- Set up apparatus far from the edge of the desk.
- Use knives or other sharp-pointed instruments with care.
SAFETY PRECAUTIONS

• Pull plugs, not cords, when removing electrical plugs.
• Clean glassware before and after use.
• Check glassware for scratches, cracks, and sharp edges.
• Clean up broken glassware immediately.
• Do not use reflected sunlight to illuminate your microscope.
• Do not touch metal conductors.
• Use alcohol-filled thermometers, not mercury-filled thermometers.

USING CHEMICALS

• Never taste or inhale chemicals.
• Label all bottles and apparatus containing chemicals.
• Read labels carefully.
• Avoid chemical contact with skin and eyes (wear safety glasses, lab apron, and gloves).
• Do not touch chemical solutions.
• Wash hands before and after using solutions.
• Wipe up spills thoroughly.

HEATING SUBSTANCES

• Wear safety glasses, apron, and gloves when boiling water.
• Keep your face away from test tubes and beakers.
• Use test tubes, beakers, and other glassware made of Pyrex™ glass.
• Never leave apparatus unattended.
• Use safety tongs and heat-resistant gloves.
• If your laboratory does not have heat-proof workbenches, put your Bunsen burner on a heat-proof mat before lighting it.
• Take care when lighting your Bunsen burner; light it with the airhole closed, and use a Bunsen burner lighter in preference to wooden matches.
• Turn off hot plates, Bunsen burners, and gas when you are done.
• Keep flammable substances away from flames and other sources of heat.
• Have a fire extinguisher on hand.

FINISHING UP

• Thoroughly clean your work area and any glassware used.
• Wash your hands.
• Be careful not to return chemicals or contaminated reagents to the wrong containers.
• Do not dispose of materials in the sink unless instructed to do so.
• Clean up all residues and put them in proper containers for disposal.
• Dispose of all chemicals according to all local, state, and federal laws.

BE SAFETY CONSCIOUS AT ALL TIMES!