



## Which Orange Juice Has the Most Vitamin C?

[https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem\\_p044/chemistry/which-orange-juice-has-the-most-vitamin-c](https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c) ([http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem\\_p044/chemistry/which-orange-juice-has-the-most-vitamin-c](http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c))

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### Experimental Procedure

**Caution:** Iodine solution is poisonous. Avoid skin and eye contact. See the [Materials Safety Data Sheet](https://www.homesciencetools.com/content/reference/CH-IODINE.pdf) (<https://www.homesciencetools.com/content/reference/CH-IODINE.pdf>) for complete information. Wear chemical safety goggles and rubber gloves when handling the concentrated solution. Iodine solution will stain clothing; a lab coat or apron is also recommended. For more information on how to properly use a balance and different titration techniques see [Chemistry Lab Techniques](http://www.sciencebuddies.org/science-fair-projects/references/chemistry-lab-techniques) (<http://www.sciencebuddies.org/science-fair-projects/references/chemistry-lab-techniques>).

1. Do your background research so that you are knowledgeable about the terms, concepts, and questions, in the [Background](http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#background) ([http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem\\_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#background](http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#background)) section. For information about doing a titration, visit the Science Buddies webpage [Titration Tutorial: Tips & Tricks for Titrating](http://www.sciencebuddies.org/science-fair-projects/references/titration-tutorial-tips-tricks-for-titrating) (<http://www.sciencebuddies.org/science-fair-projects/references/titration-tutorial-tips-tricks-for-titrating>).
2. Wear gloves, chemical safety goggles, and a lab coat or apron when using the iodine solutions in this experiment. Also, if you are not working on a surface that can be stained, you should completely cover the surface with newspaper to protect it.
3. Dilute the Lugol's iodine solution 1:10 in distilled water to make your iodine titration solution.
  - a. Carefully pour 30 milliliters (mL) of Lugol's solution into the 500 mL graduated cylinder. This amount should be the entire bottle that comes with the Science Buddies kit.
  - b. Add enough distilled water to bring the total fluid volume to 300 mL and mix, as shown in Figure 1, below.



**Figure 1.** After adding the 30 mL of Lugol's iodine solution to the 500 mL graduated cylinder, add enough distilled water to bring the total fluid volume up to 300 mL.

- c. Slowly and carefully pour the diluted solution into an amber glass bottle. You should use a funnel to do this.
  - i. **Caution:** The amber glass bottles that come with the Science Buddies kit hold about 230 mL each. This means you will need to use *both* bottles to store the 300 mL of solution. Be careful when filling the first bottle so that it does not overflow!
- d. Label the bottle(s), such as by using a permanent marker and masking tape.
- e. You can store the solution in its tightly sealed bottle(s) for now.
  - i. *Note:* Iodine is light-sensitive. Storing the solution in an amber glass bottle (or an aluminum foil-covered bottle), or in a dark location, protects it from light.
- f. Rinse and dry the 500 mL graduated cylinder.

4. Make a starch indicator solution. This can be anywhere from 0.5 to 1.0%. The exact amount of starch is not critical.
  - a. Heat 200 mL of distilled water in a pot on the stove.
  - b. When it is near boiling, for a 0.5% solution, add 1 gram (g) (which is equivalent to 1/4 teaspoon) of soluble starch to the pot.
  - c. Stir to dissolve the starch, while keeping the water near boiling.
    - i. Because even "soluble" starch is not very soluble, it may take about 15 minutes or more of stirring the starch to dissolve it in the near-boiling water.
  - d. After the starch is dissolved in the water, remove the pot from the burner and allow the solution to cool.
  - e. When cool, store the starch solution (at room temperature) in a clean, tightly covered glass jar that is clearly labeled.
  - f. Rinse and dry the 500 mL graduated cylinder.
5. Make a fresh vitamin C standard solution (at a concentration of 1 milligram [mg] per milliliter, or 1 mg/mL). Do this on each day that you measure the vitamin C in your orange juice samples.
  - a. You will use this solution to "standardize" your iodine titration solution. You will measure how much of your iodine solution it takes to oxidize a known amount of vitamin C. You can then use your iodine titration solution to determine the amount of vitamin C from test samples of different orange juices.
  - b. Using a cutting board and a knife, crush a 250 mg vitamin C tablet into powder and carefully transfer all of the powder to a bowl, as shown in Figure 2, below.



**Figure 2.** Crush the vitamin C tablet into powder and then transfer all of it to a bowl.

- c. Dissolve the powdered vitamin C tablet in 100 mL of distilled water, as shown in Figure 3, below.



**Figure 3.** Dissolve the powdered vitamin C tablet in 100 mL distilled water.

d. Pour the dissolved Vitamin C into the cleaned and dried 500 mL graduated cylinder and add distilled water to bring the total volume to 250 mL, as shown in Figure 4, below. You now have a vitamin C standard solution at a concentration of 1 mg/mL.



**Figure 4.** Carefully transfer the 100 mL of dissolved vitamin C into the 500 mL graduated cylinder and fill it with distilled water until the total volume is 250 mL.

6. Set up the 50 mL buret on the ring stand, as shown in Figure 5, below.
  - a. You will first need to assemble the ring stand from the Science Buddies kit. To do this, screw the metal rod into the black metal base. Then tightly screw the buret clamp into position on the top of the ring stand's metal rod, as shown in Figure 5.
  - b. To set up the 50 mL buret on the ring stand, carefully slide it into place between the prongs of the buret clamp, as shown in Figure 6, below.

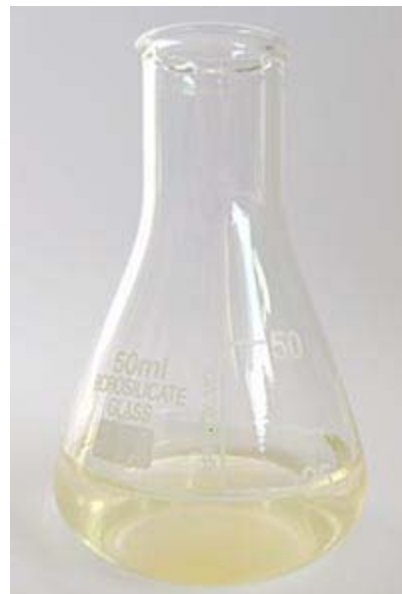


**Figure 5.** Assemble the ring stand, buret clamp, and buret, as shown here.



**Figure 6.** Carefully slide the buret into place between the prongs of the buret clamp.

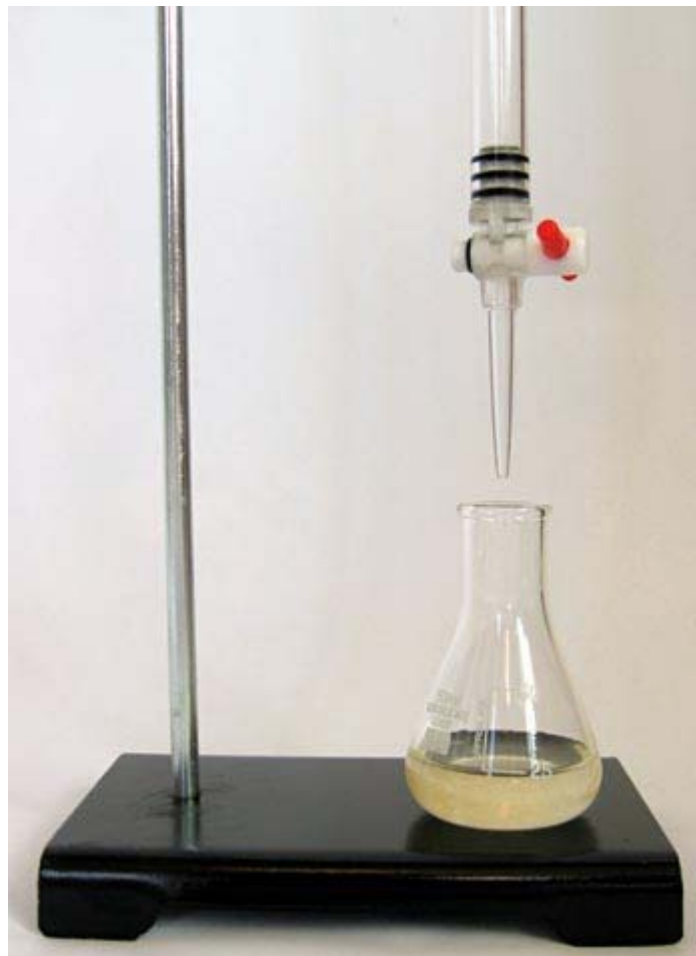
7. Titrate 20 mL of vitamin C standard solution.
  - a. Use the 50 mL graduated cylinder to measure 20 mL of vitamin C standard solution.
  - b. Pour this into the 50 mL Erlenmeyer flask, as shown in Figure 7, below. The shape of this flask allows you to swirl the solution to mix it without spilling.



**Figure 7.** Pour 20 mL of the vitamin C solution into the 50 mL Erlenmeyer flask.

- c. Add 10 drops of starch indicator solution to the Erlenmeyer flask.
- d. Carefully adjust the position of the buret on the ring stand so that you can place the Erlenmeyer flask (containing the vitamin C and starch solutions) right under the buret, as shown in Figure 8, below. Leave the Erlenmeyer flask there.





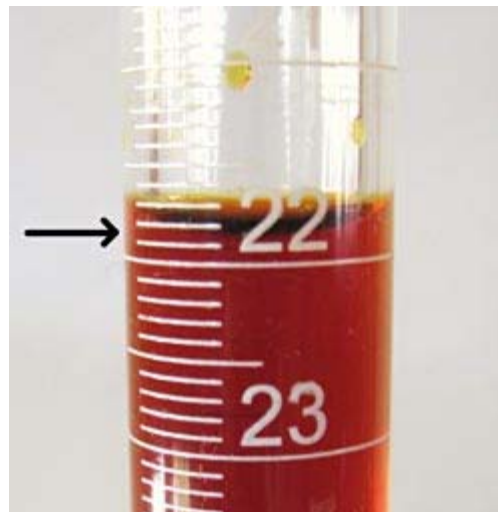
**Figure 8.** Adjust the buret's position so you can place the 50 mL Erlenmeyer flask right underneath it.

- e. Make sure the bottom of the buret is turned to the closed position. This is done by turning the red stopper to the horizontal position, as shown in the close-up in Figure 9, below, for the buret from the Science Buddies kit.



**Figure 9.** Make sure the buret is turned to the closed position, as shown here with the buret from the Science Buddies kit.

- f. Use the funnel to carefully fill the buret with the diluted Lugol's iodine solution you prepared in step 3. Fill it somewhere between the 5 mL and 35 mL marks on the buret. The exact position is not important, as long as the fluid level is not past the graduated markings on the top of the buret.
- g. To make sure your measurements are accurate, fill the tip of the buret with the diluted Lugol's iodine solution. To do this, put an extra beaker below the buret and slowly turn the red stopper at the bottom of the buret (to the vertical position) to let a few drops of titrating solution flow into the beaker (or just let enough solution flow so that the entire tip of the buret is full of solution). Then, making sure the bottom of the buret is closed again, place the Erlenmeyer flask back under the buret.
- h. In your lab notebook, write down the initial level (in mL) of the iodine solution in the buret. Read the level from the bottom of the *meniscus*, which is the curved surface of the liquid.
  - i. For example, in Figure 10, below, the level should be recorded as 21.85 mL, since this is where the bottom of the meniscus is.



**Figure 10.** When reading the level of liquid in the buret, read from the bottom of the meniscus, which is being pointed to with a black arrow in this picture. For example, the level of the liquid in this buret should be read as 21.85 mL. (*Note:* The long white line at the top of the buret is the mark for 21 mL.)

- i. Slowly turn the red stopper at the bottom of the buret until just one drop of the iodine solution comes out into the Erlenmeyer flask below it. Then close the buret again.
  - i. *Note:* It is important to only let the iodine solution be added one drop at a time because the titration reaction is very sensitive. One drop can be enough to drive the reaction to completion (if it were near completion before). If more than one drop is added at a time, the data will not be as accurate as it could be.
- j. After each drop is added, swirl the flask to mix in the iodine solution.
  - i. When adding the iodine solution, you may see a temporary color change that goes away when you swirl the flask, as shown in Figure 11, below.



**Figure 11.** When you add a drop of iodine solution, you may see a temporary color change, as shown here (the dark blue swirls), that goes away when the iodine solution is mixed in the flask by swirling it.

- k. Continue adding the iodine solution, mixing in one drop at a time and swirling the flask, until the iodine creates a blue-back color throughout the solution, as shown in Figure 12, below, that lasts for longer than 20 seconds. At this point, the titration is complete.



**Figure 12.** When the entire solution in the flask changes to a blue-black color that lasts for more than 20 seconds while swirling it, the titration is complete.

- l. In your lab notebook, record the final level of the iodine solution remaining in the buret.
  - m. The difference between the initial level and the final level is the amount of iodine titration solution needed to oxidize the vitamin C.
  - n. Rinse out and dry the Erlenmeyer flask.
  - o. Repeat step 7 three times. You should get results that are within about 0.2 mL of each other. If you do not, repeat this step until you have three results that are within about 0.2 mL of each other.
    - i. Be sure to check the level of the iodine solution in the buret before each trial. Remember, if the level is below the 35 mL mark, carefully add more iodine solution to the buret (until it is between the 5 mL and 35 mL marks). Record the new level.
8. Prepare fresh-squeezed orange juice for testing.
- a. Use the juicer to extract orange juice from three (or more) oranges, as shown in Figure 13, below.
  - b. You need 20 mL of juice per titration, and you should do at least three titrations of the fresh-squeezed orange juice, so you will need at least 60 mL total.
  - c. Filter the orange juice through cheesecloth to remove any pulp and seeds.



**Figure 13.** Use a juicer, like the one shown here from the Science Buddies kit, to juice at least three oranges. You will need to cut each orange in half to use this juicer.

9. Titrate the fresh-squeezed orange juice you just prepared by repeating step 7, but this time, use 20 mL of fresh-squeezed orange juice in the Erlenmeyer flask instead of 20 mL of the vitamin C solution.
  - a. Be sure to check the level of the iodine solution in the buret before each trial. Remember, if the level is below the 35 mL mark, carefully add more iodine solution to the buret (until it is between the 5 mL and 35 mL marks). Record the new level.
  - b. In step 7.k., the titration is complete when the iodine creates a distinct color change in the juice/starch solution. This color change will be harder to see than with the vitamin C solution, since the juice starts out orange. The color will change from orange to grayish brown when the endpoint is reached, as shown in Figure 14, below. If you continue to add iodine, the color will darken further, but you want to note the volume of iodine added when the color *first* changes.
  - c. Remember to record the final level of the iodine solution in the buret, as done in step 7.i.
  - d. The difference between the initial level and the final level is the amount of iodine titration solution needed to oxidize the vitamin C in the orange juice.
  - e. As you did for step 7.o., repeat the titration process for the fresh-squeezed orange juice a total of three times. You should get results that are within about 0.2 mL of each other.



**Figure 14.** When the titration is complete with the orange juice sample, the entire orange juice sample will turn from orange to grayish brown in color, as shown here.

10. Titrate your other orange juice samples by repeating step 9 using the premium not-from-concentrate orange juice and the orange juice made from frozen concentrate (separately).
  - a. If you test the orange juice samples on different days, be sure to make a fresh vitamin C standard solution on each day that you measure the vitamin C in your orange juice samples.
11. For each juice (fresh, premium, or from-concentrate), calculate the average amount of iodine needed to titrate the 20 mL sample. Record your results in your lab notebook.
12. Calculate the amount of vitamin C in your samples by setting up a proportion, as shown in Equation 1, below. You will want to solve for Vitamin C<sub>2</sub>

**Equation 1.**

$$\frac{\text{Iodine}_1}{\text{Vitamin } C_1} = \frac{\text{Iodine}_2}{\text{Vitamin } C_2}$$

- Iodine<sub>1</sub> is the average amount of iodine (in mL) needed to titrate the vitamin C standard solution.
- Vitamin C<sub>1</sub> is the amount of vitamin C in the standard solution (in mg).
- Iodine<sub>2</sub> is the average amount of iodine (in mL) needed to titrate the orange juice sample.
- Vitamin C<sub>2</sub> is the amount of vitamin C in the orange juice sample (in mg).

- a. You can rearrange Equation 1 to directly solve for the unknown "Vitamin C<sub>2</sub>" value, resulting in Equation 2, below.

- i. If you are not sure how Equation 2 was derived from Equation 1, take a moment to examine the equations so that you understand the derivation process.

**Equation 2.**

$$\text{Vitamin } C_2 = \frac{\text{Iodine}_2 \times \text{Vitamin } C_1}{\text{Iodine}_1}$$

- b. Here is an example (with sample numbers) to show you how to use Equation 2:
- Let us say that it took an average of 8.5 mL of iodine solution to titrate 20 mL of 1 mg/mL vitamin C standard solution. 20 mL of the vitamin C solution is equal to 20 mg vitamin C total. This means that "Iodine<sub>1</sub>" equals 8.5 mL and "Vitamin C<sub>1</sub>" equals 20 mg.
  - Let us also say it takes an average of 6.8 mL of iodine solution to titrate a 20 mL test sample of orange juice. This means that "Iodine<sub>2</sub>" equals 6.8 mL.
  - Using this example, we would set up Equation 2 to look like Equation 3, below. Note that we do not know "Vitamin C<sub>2</sub>" because that is what we are solving for.

**Equation 3.**

$$\text{Vitamin } C_2 = \frac{6.8 \text{ mL} \times 20 \text{ mg}}{8.5 \text{ mL}}$$

- iv. When solving for Vitamin C<sub>2</sub> in Equation 3, we get an answer of 16 mg. This means there is 16 mg of vitamin C in that orange juice sample.
13. Graph your results, putting the names of the different kinds of orange juice on the x-axis and the average amount of vitamin C on the y-axis.
14. Look at your graphs. Did one type of orange juice have more vitamin C than the others? Why do you think this might be? Can you explain your results?

## Frequently Asked Questions (FAQ)

FAQ for this Project Idea available online at [https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem\\_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#help](https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#help) ([http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem\\_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#help](http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p044/chemistry/which-orange-juice-has-the-most-vitamin-c#help)).