Determination of Vitamin C in Foods

Object

The object of this experiment is to measure the amount of ascorbic acid in common foods using an oxidation reduction reaction.

Introduction

Vitamin C, has the chemical name ascorbic acid. It is a water soluble vitamin. Although it is important for good health, humans do not have the ability to make their own vitamin C and must obtain it through diet or take it in vitamin supplements. Citrus fruits, potatoes and some green vegetables are known to be good sources of vitamin C. (See Table 1) Plants synthesize the compound for the growth, development, and protection of the plant. The exact pathway for its synthesis is not well understood.

Plant source	Amount of Vitamin C (mg / 100g)		Plant source	Amount of Vitamin C (mg / 100g)
Rose hip	426		Spinach	30
Red pepper	190		Potato	20
Parsley	130		Green Beans	16
Broccoli	90		Tomato	10
Brussels sprouts	80		Watermelon	10
Elderberry	60		Banana	9
Strawberry	60		Carrot	9
Orange	50		Apple	6
cantaloupe	40		Lettuce	4
grapefruit	30		Raisin	2

Table 1. Amount of Vitamin C in Various Plants

Vitamin C is notoriously easy to destroy. Orange juice may lose half of its vitamin C in a week in the refrigerator, but cut fruit may lose much less after 6 days (Some examples: mango, strawberry, and watermelon: less than 5%, Pineapple: 10%, Kiwi: 12%, and Cantaloupe: 25%, after 6 days). Cooking also destroys vitamin C. The US Department of Agriculture reports the following vitamin C levels:

Amount of vitamin C, mg/100g			
Vegetable	**Raw**	**Frozen**	**Frozen then boiled**
Green beans	12.2	9.7	4.1
Broccoli	89.2	64.9	40.1

Note: Freezing fresh vegetables requires blanching, i.e., boiling for a short period to stop enzyme actions in the plant.

Some cooks had a practice of adding sodium bicarbonate, NaHCO3, to vegetables to maintain a bright green color. That practice essentially destroys the Vitamin C in the vegetables.

Vitamin C is required for the synthesis of collagen, an important structural component of blood vessels, tendons, ligaments and bone. It also is important in the synthesis of the neurotransmitter norepinephrine, which is critical to brain function and can affect mood.

Vitamin C is also a highly effective antioxidant. In small amounts vitamin C can protect indispensable molecules in the body, such as proteins, lipids (fats), carbohydrates, and nucleic acids (DNA and RNA), from damage by free radicals and reactive oxygen species that can be generated during normal metabolism as well as through exposure to toxins and pollutants. It has been recommended that vitamin C, taken in large quantities on a daily basis, may prevent the common cold. Some recommended dietary allowances for vitamin C are listed in Table 2.

Age	**Male**	**Female**	**Pregnancy**	**Lactation**
0–6 months	40 mg*	40 mg*		
7–12 months	50 mg*	50 mg*		
1–3 years	15 mg	15 mg		
4–8 years	25 mg	25 mg		
9–13 years	45 mg	45 mg		
14–18 years	75 mg	65 mg	80 mg	115 mg
19+ years	90 mg	75 mg	85 mg	120 mg
Smokers	Individuals who smoke require 35 mg/day more vitamin C than nonsmokers.			

Table 2. Recommended Dietary Allowances (RDAs) for Vitamin C

Ascorbic acid is the chemical name for vitamin C. As the name indicates vitamin C is an acid. Therefore, a base such as sodium hydroxide can neutralize it. Ascorbic acid is also easily oxidized. Both of these reactions can be used for quantitative analysis of the compound.

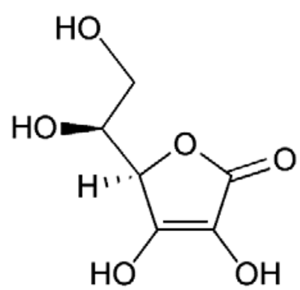

Figure 1. The structure of ascorbic acid, $C_6H_8H_6$
(MW = 176.12 g/mol)

In this experiment, you are performing an oxidation-reduction titration. You will first determine the concentration of the iodine solution by titrating a solution of ascorbic acid. Next, you will titrate fruit juices or a fruit or vegetable extract. Ascorbic acid reacts with iodine through the following reaction:

$$C_6H_8O_6 + I_2 \rightarrow C_6H_6O_6 + 2\, I^- + 2\, H^+$$

The equivalence point of the titration will be determined using a starch indicator. Molecular iodine combines with starch to make a dark blue complex. In the titration reaction, molecular iodine is added in the form of the iodine solution. In the presence of ascorbic acid, the iodine is reduced to colorless iodide ion which does not react with starch to make a colored complex. When all of the ascorbic acid has been titrated, any additional iodine added, present in the solution as molecular iodine, will combine with the starch to make the solution blue. The appearance of the blue solution will signal the equivalence point. As with the acid-base titration, the iodine solution must be standardized against a known solution of ascorbic acid, which you will prepare from the solid.

Safety

Goggles or safety glasses must be worn at all times in the laboratory.

The iodine solution is an irritant and will stain the skin and clothing. The solution does give off a small amount of iodine vapors. Iodine vapors are toxic by inhalation. Work in a well ventilated area. Keep the iodine solution covered.

Disposal

Dispose of all solutions in the waste containers provided.

Materials Needed

Ascorbic Acid
Iodine Solution
2% Starch solution (indicator)
125 ml Erlenmeyer flasks
Buret
Magnetic stirrer with stir bar
Small funnel for filling buret
Blender
Knife
Small cutting board (wood or plastic)
Cheesecloth
Funnel
Scissors
Optional: gloves

Experimental Procedure

Standardization of the iodine solution

You will use a solution of ascorbic acid to standardize the iodine solution.

1. Weigh three samples of solid ascorbic acid, approximately 0.05g to 0.10 g each.

2. Place each sample in a numbered 125 ml Erlenmeyer flask. Add about 30ml distilled water and 4 to 5 drops of starch solution to each flask.

3. Rinse and fill a 25 or 50 mL buret with the iodine solution and titrate the ascorbic acid solution. Be sure to record the initial and final volume readings from the buret.

4. After the data is collected, calculate the molarity of the iodine solution.

5. Repeat the procedure for the other two ascorbic acid solutions

Determination of Vitamin C in a Fruit Juice or Food

Each group will run two sets of titrations, one with fresh fruit juice and refrigerated juice (e.g., juice from an orange and packaged orange juice) OR one with a fresh vegetable and one with frozen vegetable (e.g., broccoli and frozen broccoli).

Preparation of samples

For fresh fruit juice from oranges, grapefruits, or lemons.

If using fresh fruit, squeeze at least 100 mL of the juice from the fruit. If the juice contains pulp and any seeds, strain 100 mL of juice through cheesecloth.

For packaged fruit juice.

If the juice contains pulp, strain 100 mL of juice through cheesecloth.

For fresh or frozen fruits and vegetables.

Cut a 100 g sample into small pieces.

Place the sample into a blender or food processor, add 50 mL distilled water and blend to a pulp.

Strain the fruit or vegetable pulp through cheesecloth, washing the pulp with a few 10 mL portions of water and collecting all filtrate in a 150-mL or 250-mL beaker.

Pour the filtrate into a 100 mL graduated cylinder and dilute the solution to 100 mL with distilled water.

Titration of the fruit juice or extract

1. Measure 20 mL of your sample solution into a 125 mL Erlenmeyer flask. Add 25 mL of distilled water and 1 mL of starch indicator solution.

2. Titrate the sample with your standardized iodine solution from the first part of this experiment. The endpoint of the titration occurs when you obtain a permanent dark blue-black color due to the starch-iodine complex.

3. Repeat the titration twice more with additional 20 mL aliquots of your sample solution. Your titration results should agree to within 0.5 mL. If they do not agree, see your instructor. You may have to do an additional titration.

4. If you started with juice or extract from fresh fruit or vegetable, in step 1, above, repeat the series of titrations using samples prepared from the packaged juice or extract from frozen vegetables. If you

stared with the packaged juice and frozen vegetable, then use samples prepared from the fresh juice or vegetable.

5. Determine the amount of ascorbic acid present in the samples of fruit juice or vegetable you titrated. Compare your result to the vitamin C content listed on the label of the packaged juice, from Table 1 in this experiment, or from data obtained on the Internet.

Determination of Vitamin C in Foods

Data and Results

Name____ Course/Section ______________________

Partner's Name (If applicable) ____ Date _______________

Standardization of the iodine solution

Mass of ascorbic acid used, g	Initial Volume of Iodine Solution, mL	Final Volume of Iodine solution, mL	Volume of Iodine Solution used, mL	Concentration of Iodine Solution M

Formula for M_{iodine}:

$$M_{Iodine} = mass_{ascorbic\,acid} \times \frac{1\,mole_{ascorbic\,acid}}{176.12\,g_{ascorbic\,acid}} \times \frac{1000\,mL/L}{Volume_{Iodine\,solution,\,mL}}$$

Sample calculation for M_{iodine} (Show the set-up for one calculation)

Average concentration of iodine solution ______________________ M

Determination of Vitamin C in a Fruit Juice or Food

Fruit or fruit juice used ____

Mass of fresh fruit or vegetable OR volume of fresh fruit juice used ________________________

Volume of fruit juice or extract used, mL	Initial Volume of Iodine Solution, mL	Final Volume of Iodine solution, mL	Volume of Iodine Solution used, mL	Concentration of Ascorbic Acid in Sample mg

Mass of frozen fruit or vegetable OR volume of packaged fruit juice used _____________________

Volume of fruit juice or extract used, mL	Initial Volume of Iodine Solution, mL	Final Volume of Iodine solution, mL	Volume of Iodine Solution used, mL	Concentration of Ascorbic Acid in Sample mg

Formula for Concentration of Ascorbic Acid in your sample:

$$mg_{ascorbic\,acid} = M_{iodine\,solution} \times mL_{iodine\,solution} \times 176.12\, g/mole$$

Sample calculation for $mg_{ascorbic\ acid}$ (Show the set-up for one calculation)

Summary of titration results

Average concentration of Ascorbic Acid in fresh fruit or vegetable samples ______________________ mg

Average Volume of fresh fruit juice or extract used for each titration ______________________ mL

Average concentration of Ascorbic Acid in packaged fruit or frozen vegetable samples ______________________ mg

Average Volume of packaged fruit juice or frozen vegetable extract used for each titration ______________________ mL

Concentration of Ascorbic acid in Food

(Note: This equation gives answer in mg Ascorbic acid/100 g fruit. We are assuming that 1 mL of fruit juice or fruit extract has a mass of 1 g.)

$$\text{Concentration}_{\text{Ascorbic Acid}} \text{ in fruit used} = \frac{\text{Average Conc.}_{\,Ascorbic\,acid}}{\text{Average Volume}_{Fruit\ juice}} \times \frac{1\,mL_{Fruit\ juice}}{1\,g_{Fruit\ juice}} \times 100$$

Sample calculation for $\text{Concentration}_{\text{ascorbic acid}}$ (Show the set-up for one calculation)

Final Results

Average concentration of Ascorbic Acid in fresh fruit juice or vegetable tested ___________________ mg/100 g

Average concentration of Ascorbic Acid in packaged fruit juice or frozen vegetable tested ___________________ mg/100 g

Questions

1. What is the concentration of vitamin C in the juice or fruit tested according to the package label, Table 1 (in the experiment), or from Internet data?

2. How does the concentration of vitamin C from your result of this experiment compare to the values in question 1, above?

3. Can you explain any differences you observed in question 2, above?

4. Your group tested fresh fruits, vegetables, or juices, and frozen or packaged fruits, vegetables, or juices, how does their vitamin C concentrations compare?

5. Do you have any recommendations for buying, cooking and storing juices, fruits, and vegetables?