

Beta-Carotene Extractions

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 Subject: Biology
 Level: High School Biology Students
 Standards: *New York State*(www.emsc.nysed.gov/ciai/)
 Standard 1- Analysis, Inquiry and Design
 Standard 4- Matter is made up of particles whose
 properties determine the observable characteristics
 of matter and its reactivity.
 Standard 7- Interdisciplinary Problem Solving

Schedule: Five 50 minute class periods

Objectives:

Examine molecular structure and properties of Beta-carotene. Determine amounts of Beta-carotene in sweet potato due to culinary process (baked, boiled, and fried).

Students will:

- Differentiate between cancer prevention and cancer therapy.
- Identify the relationship between cancer and sweet potatoes.
- Illustrate the different culinary processes of sweet potatoes (baked boiled and fried).
- Measure culinary ingredients using English-metric conversion system.
- Demonstrate the extraction process of B-carotene in sweet potatoes.
- Review and discuss concepts learned throughout activity.

Vocabulary:

Beta-carotene	Spectroscopy
Column	Vacuum filtration
Chromatography	TLC
Extraction	UV

Materials:

For Each Pair:	Hot plates
Weigh boat	Cooking Utensils
Acetone	Heat Mitt
Vacuum filter	Water
Test tubes	Butter/Oil
CH ₂ Cl ₂	Cinnamon/Sugar
Cooked Sweet	Para film
Potatoes	Refrigerator
CaCl ₂ pellets	Hexane
Chemical hood	Vials
Vials with caps	Sand
Funnel	Cotton
Filter paper	Silicone gel
Filter	Pipette
Graduated cylinder	Graduated cylinder
Beakers (various sizes)	Silica gel plates
Prepared slide plates	syringe
	Spect 20's

Safety:

Students should use extreme caution when using laboratory glassware handling chemicals and should always wear protective safety gear.

Science Content for the Teacher:

Pre-Teaching Concepts:

- Beta- carotene is responsible for the orange color in foods.
- Sweet potatoes are rich in beta-carotene.
- Students must think through the difference between cancer prevention and cancer therapy.
- Various extraction methods.
- Difference between TLC and column chromatography.

What is an extraction? A method used to separate compounds based on their relative solubilities.

Liquid-liquid extraction is an extraction of a substance from one liquid phase into another liquid.

Preparation:

1. Purchase culinary materials or have students bring materials.
2. Photocopy print materials (Activity Sheets and Lab Activity 1-4) for each student group.
3. Distribute materials evenly to each student pair or group.



Classroom Procedure:

Day One

1. Discuss the basic concepts of light outlined in 'Science Content' with the students. Demonstrate the 'Pre-Teaching Concepts' and discuss the terminology.
2. Students are separated into groups to determine individual roles. Students should decide which group members are responsible for specific tasks throughout the activity.
3. Students should begin discussing the different types of sweet potato culinary processes and the effect they have on nutrition. Students will also discuss the environmental factors of sweet potatoes and how they assist in cancer prevention.
4. Students should begin working on Activity Sheet 1. Allow enough time for students to complete this.
5. Allow time for questions on the concepts learned.
6. Encourage students to review the concepts they learned and to ask any questions they may have before the following class period.

Day Two

1. Students should collect all necessary materials for the day's activities.
2. Review the concepts learned during the previous class period and answer any questions they may have.
3. Students should begin working on Lab Activity 1: Sweet potato preparation. Encourage the groups to try to complete as much of the sweet potato preparation as possible. Assist as necessary.
4. Ask students to clean up their workstations and dispose of all materials.
5. Allow time for a question and answer session. Review and discuss the concepts learned during the day's activities.

Day Three

1. Students should collect all necessary materials for the day's activities.
2. Students should begin working on Lab Activity 2: Extraction of Beta-carotene. Encourage the groups to try to complete most of the lab activity. Assist as necessary.
3. Encourage discussion within groups once the students begin working on Activity Sheet 2.
4. Ask students to clean up their workstations and dispose of all materials.
5. Allow time for a question and answer session. Review and discuss the concepts learned during the day's activities.



Day Four

1. Students should collect all necessary materials for the day's activities.
2. Students should begin working on Lab Activity 3: Purification with TLC- Thin Layer Chromatography and Column Chromatography. Encourage the groups to try to complete most of the lab activity. Assist as necessary.
3. Encourage discussion within groups once the students begin working on Activity Sheet 3.
4. Ask students to clean up their workstations and dispose of all materials.
5. Allow time for question and answer session. Review and discuss the concepts learned during the day's activities.

Day Five

1. Students should collect all necessary materials for the day's activities.
2. Students should begin working on Lab Activity 4: Characterization using UV spectroscopy. Encourage the groups to try to complete most of the lab activity. Assist as necessary.
3. Encourage discussion within groups once the students begin working on Activity Sheet 4.
4. Ask students to clean up their workstations and dispose of all materials.
5. Allow time for a question and answer session. Review and discuss the concepts learned during the day's activities. Answer any questions the students may have regarding the principles they learned.



Safety:

- Students should always use caution in the lab when handling hot objects and chemicals. Always wear safety clothing and equipment while working in the laboratory.



Lab Methods & Instructions

Lab Activity 1: Sweet Potato Culinary Process

- ❖ Students should begin with preparation of sweet potatoes (bake, boil, and fry). Peel and chop at least two sweet potatoes first. Use those potatoes for boiling and frying (Use oil to fry the potato). The third sweet potato will be used for baking. To boil, add 200 ml of distilled water to cover the potatoes. To fry, add 1 tbsp of cooking oil to pan, cook potato on medium heat until potato is done (Heat may be adjusted). To bake, place potato in a pre-heated 375^o oven. Bake for at least one hour. When boiled sweet potatoes have finished cooking, add 1 tbsp of butter, 12g of sugar and 2g of cinnamon. For the fried sweet potato add 2g of sugar and cinnamon.
- ❖ Place each cooked sweet potato in a separate beaker. Mash sweet potato with a pestle until potato is soft. In a graduated cylinder measure 20 ml of acetone to add to potato. Continue measuring and adding acetone until potato is saturated. Be sure to count of the amount of acetone being added. Once sweet potatoes have been saturated, weigh the content and record it. Then cover sweet potatoes with para film and store in the refrigerator over night.

Lab Activity 2: Extraction of Beta-carotene

- ❖ Using a pipette, extract all aqueous layers from potatoes and discard them. This is a very slow process and may have to be repeated several times, but be sure to remove any excess water from the sweet potatoes. Using a weigh boat, weigh the content and record it.
- ❖ Using a filter funnel, filter paper and an Erlenmeyer flask, begin filtering the sweet potato substances. Before placing the filter paper in the funnel, saturate the filter paper with acetone. Place the filter paper in the funnel and place the funnel in the flask. Remove all sweet potato substance from the beaker and place it in the funnel. This will filter all water from the sweet potato through gravity filtration. Once all liquid substance has been removed, place the remaining solid substances in another beaker.
- ❖ Add 20 ml of acetone to the solid substance. Measure out 15g of the substance; add 2 ml of acetone and 15 ml of CH₂Cl₂, then place substance in another funnel, this time use a rubber stopper to hold the funnel in place. Attach a rubber hose to a flask with an open side. Attach the other end of the hose to the water faucet handle. Place rubber attached funnel inside the flask. Turn water on, the pressure from the water will suction the excess liquid from the sweet potato substance.



- ❖ This process is called vacuum filtration. Remove CH_2Cl_2 into a separate tube and add CaCl_2 pellets (2, 2-3 drops). Remove any liquid and place it in a test tube. Next drive off CH_2Cl_2 directly under the chemical hood.
- ❖ Weigh vials and sub heat. Add 80/20 (80-hexane and 20 acetone) to vials. Re-dissolve hexane (try to get all into the solution). Fill glass jar with 80/20 solution and place lid on tight (this can be used for the following day procedure).

Lab Activity 3:

Purification with TLC-Thin Layer Chromatography

- ❖ Before beginning draw a $\frac{1}{2}$ inch line from the top and from the bottom of each sample plate. Using a syringe, remove a drop of substance from each vial and place it on the silica gel plate. Place the plate sample in the glass jar filled with the 80/20 solution and replace the lid. The solution will cause the evidence of B-carotene (if present) to appear. Record your results and discard the solution in the jar.

Purification via Column Chromatography

- ❖ Place a small piece of cotton, a layer of sand, a layer of a mixture of silicone gel and hexane in a glass pipette. Pump air to content, and then mix more sand to seal off 80/20 hexane mixture in the column. Pour B-carotene substance, add hexane solution; make sure that you do not allow the hexane solution to run out. Capture the B-carotene substance and hexane solution in a test tube, set aside. This will be used in the next activity.

Lab Activity 4: Characterization using UV spectroscopy

- ❖ Place the previous activities substances on a silica gel plate sample. Using a syringe, remove a drop of substance from each test tube and place the drop on the silica gel plate. Using the Spec 20, notice if any evidence of B-carotene is present. Record all data whether evidence was present or not.



Activity Sheet 1

Sweet Potato Culinary Process

Questions:

1. Are there any healthy sweet potato recipes?
2. What are some of the nutritional concerns about sweet potatoes?
3. What relationship does cancer and sweet potatoes have?
4. How does Beta-carotene aid in cancer prevention?

Homework

What are some ways to extract nutrients from food?



Activity Sheet 2

Extraction of Beta-carotene

Questions:

1. What are the procedures for extracting Beta-carotene?

2. How does Beta-carotene aid in cancer prevention?

Homework

Define the word purification.



Activity 3

Purification with TLC-Thin Layer Chromatography and Column Chromatography

Questions:

1. What is the process of TLC and do we use it?
2. How is this type of chromatography different from TLC? How are they similar?
3. Why is chromatography so important in this type of research?



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