Science Experiment:
The Brown Apple Project
Project: Food Science, Plant Science

Objectives:
Participants will explore the chemical reaction behind apple browning.
Time to complete activity: 1 hour and 15 minutes

Background:
Fruit turns brown when exposed to air because a reaction is happening when a cut piece of fruit is exposed to oxygen. This is calledenzymatic browning. The name enzymatic browning comes from the fact that an enzyme located in the fruit reacts with oxygen from the air to turn the fruit brown.
The chemical reaction can be simplified to:
Polyphenol Oxidase + O_2 → Melanin (Brown Color)
Oxygen activates the compound *polyphenol oxidase* in the fruit to turn the fruit brown. *Polyphenol oxidase* is the enzyme.

- Definition of an enzyme: A substance produced by all living organisms that speed up a chemical reaction (i.e. speeding up the browning of fruit).
- FUN FACT: Enzymes usually end with the suffix *-ase.*

Materials:

- Fresh apple slices
- Lemon juice
- Water
- Fruit Fresh® (product used to prevent browning)
- Apple juice
- White vinegar
- Tongs
- Paper towels
- Paper plates
- Paper Bowls
- Clock or timer

Set Up:
There should be different areas set up for each group to perform the upcoming experiment.
Prior to starting the activity, talk about it!

- What happens when you cut into a piece of fruit and leave it on the counter?
- Fruit is an important part of our diets to keep us healthy, but who wants to eat a brown, soft piece of fruit? Most people prefer a fresh, crisp apple for instance over a mealy, mushy, and brown apple. What about you guys? Would you like to eat a brown fruit?
- Can anyone think of a way to prevent this from happening?
- Allow the kids to openly brainstorm and consider
    - Various ideas (would you stick it in the refrigerator…wait to cut the fruit until you want to eat it etc.)

Introduce the experiment:
This experiment involves dipping slices of apple in a variety of substances. These include water, lemon juice, a special substance designed to preserve fruit, and apple juice. The fruit will be observed over time (use our data sheet or make one of your own).
Before beginning, ask participants what they think will happen? Which mixture will prevent browning the most? Have the participants write down their predictions.

Methods

1. Take out 3 bowls and put water, lemon juice, and vinegar in the bowls respectively.
2. Label a paper plate "water."
3. Using the tongs dip apple slice(s) into water for 30 seconds.
4. Take out the apple slice(s) and place the paper plate labeled "water."
5. Label a paper plate "lemon juice."
6. Using tongs, place apple slice(s) into the lemon juice for 30 seconds.
7. Take out the apple slice(s) and place on the paper plate labeled "lemon juice."(make sure to rinse the tongs aver each use to avoid cross contamination!)
8. Label a paper plate "control."
9. Place apple slice(s) on this plate without dipping them into anything.
10. Place on a paper plate labeled "Fruit Fresh®."
11. Sprinkle apple slice(s) with Fruit Fresh®.
12. Using tongs dip an apple slice(s) into apple juice for 30 seconds.
13. Place on a paper plate labeled "Apple Juice." (remember to rinse the tongs!)
14. Using tongs, dip apple slice(s) in vinegar for 30 seconds.
15. Place on a paper plate labeled "Vinegar."
16. Record your observations every 10 minutes for three intervals.

Questions to discuss

- Tell me what you noticed happening in this experiment?
- Why do you think this happened?
- How did the apple slices change in their appearance?
- Which group had the least amount of browning?
- Which had the most?

Have the youth openly discuss what they think happened. You could even go up to specific groups and hold up their apples for comparison to see if everyone had the same treatment with the least and most amount of browning.

- Ask youth to identify the ingredient that prevented the apples from turning brown the most.
- Why do they think that ingredient worked the best?

What should happen?
The treatment group of apple slices that were dipped in the water should have some browning, but not as much as the control. This is due to the water restricting the amount of oxygen coming in contact with the fruit tissues. If there isn't as much oxygen available, then less reacts with PPO to convert the phenolic to melanin (brown coloring on the surface of fruits).
The control was not treated with any solution and therefore the reaction was able to proceed and the fruit turned brown.
The treatment group of apple slices dipped in the lemon juice solution should inactivate the browning enzyme. Polyphenol oxidase (the enzyme) is pH dependent. The pH of lemon juice is between 2.0-2.5. A lower pH means the substance is more acidic. The acid in lemon juice inactivates polyphenol oxidase to prevent browning.
The treatment group of apple slices sprinkled with Fruit Fresh® should prevent browning. Fruit Fresh® is a commercial product that contains vitamin C (also known as ascorbic acid). Ascorbic acid is naturally found in lemons. The vitamin C should prevent the browning reaction from occurring.

The treatment group of apple slices dipped in apple juice should prevent browning. Apple juice has a pH between 3.5-4.0. Because apple juice is less acidic than lemon juice, expect to find the apple juice didn't prevent browning as well as the lemon juice.
The treatment group of apple slices dipped in the white vinegar should prevent browning as well. The vinegar has a pH between 2.4-3.0. Vinegar should be similar to the lemon juice apples, however they could be slightly more brown.

- Can anyone remember what we said earlier happens when fruit is cut open? (Answer: the oxygen reacts with the fruit to turn it brown)
- What type of reaction is this? (Answer: Enzymatic)

Again the definition of enzymatic browning is…
Enzymatic browning a chemical reaction that occurs when the enzyme polyphenol oxidase inside of the fruit/vegetable comes in contact with oxygen.

Real Life Examples

- Apple slices at McDonalds vs. regular apples: The prepackaged apple slices are treated with sulfites that act as an antioxidant to keep the fruit crispy and from turning brown.
- When you swallow food, digestive enzymes break up the food into smaller pieces.
- Marinades for meat often contain an enzyme called papain. Papain breaks down the fiber of meat creating a more tender product.

Can anyone think of any other products where browning has been stopped?
Did you know that plants use enzymatic browning as a defense mechanism?

- Can anyone think of a reason why this is?
- Answer: When a plant is damaged, the browning of the affected area is thought to discourage animals and insects from eating the plants any further.

Why do we care about enzymatic browning?

- This is an opportunity for youth to think and discuss.

Ultimately…

1. Effect is undesirable
2. Can decrease the quality of the food/storage
3. Can lead to spoiling

Ask: Can you think of any way scientists and the food industry can prevent this reaction from occurring?
Allow for the participants to discuss their ideas openly.
To get them to think a little deeper you could say:

- Tell me about what we just said about the solutions we placed the apples in?
- What were there effects and why did _____ work the best?
- What did it inhibit?

Methods of Prevention:

1. Inactivate the enzyme. This can be done through heat, acid (enzyme activity is pH dependent)
2. Remove the essential compound, oxygen, from the product

You can mention that with the heat method, it is important to recognize that you will cook the fruit so this method is not always best

How this concept is applied in the food industry
In the food industry, a common way to apply the techniques of enzymatic browning is drying fruit. Drying fruit is the oldest known method for preserving food. Drying fruit is safe because you are removing moisture and this prevents microorganisms from growing in it. Dried fruit is also more compact and needs less storage space; keeps/travels well.
Light colored fruits (apples, apricots, peaches, pears) tend to darken during drying and storage. This process is called oxidation. Oxidation robs the fruit of flavor, color, and vitamins. To prevent this from happening, it is common to pretreat the fruit in a solution before beginning the process of drying.

Pretreatment methods:

1. An effective method is Sulfuring. Sulfuring commercially is done on fruits using sulfur dioxide gas
2. A sulfite dip can also be used. Either sodium bisulfite, sodium sulfite, or sodium meta-bisulfite can be used; use 3/4 to 1 1/2 teaspoons of sodium bisulfite per quart of water. Place sliced fruit in the mixture and soak for 5 minutes. If fruit is in halves place in the mixture for 15 minutes. Rinse lightly under cold water and place on drying trays. This can be done indoors or outdoors. Note: some people have asthmatic reactions to sulfur.
3. A fruit juice dip can also be used. Any fruit juice that is high in vitamin C is an effective pretreatment though it does not work as well as ascorbic acid (remember, ascorbic acid is pure vitamin C). Examples are orange, lemon, pineapple, and grape juice.
4. Honey dip — Many store bought dried fruits have been dipped in a honey solution. Honey dipped fruit is much higher in calories.

Adapted from: University of Maine 4-H STEM. http://umaine.edu/4h/youth/4-h-projects/science-engineering-technology/curricula/food-science/