Explorations with



Mad Beets! Getting to the Root of Fluorescence

Version 121422

Students will be able to determine where in a beet plant photosynthesis is occurring. Students will be able to describe the general anatomy of a plant. Students will be able to relate fluorescence to photosynthesis and biotechnology.

Laboratory Safety

- 1. Wear lab coats, gloves, and eye protection as required by district protocol.
- 2. Use caution with all electrical equipment such as PCR machines and electrophoresis units.
- 3. Use caution when using sharp edged tools such as a knife or razor blade.
- 4. Wash your hands thoroughly after handling biological materials and chemicals.

Materials Required

| Item | Quantity per group |
|--|--------------------|
| Microcentrifuge tubes (1.5 mL or 2.0 mL) | 4 |
| Knife or razor blade | 4 leaves |
| Beets with root and leaves (greens) | |
| Isopropanol | 1 mL |
| The Winston Fluorescence Reader with Photohood | 1 |
| Microcentrifuge tube rack | 1 |
| 100 - 1000 μL adjustable volume micropipette with tips | 1 |
| Gloves (optional) | 1 |
| Electronice balance (optional) | 1 |







Pre lab questions

| 4 | 1 A / I | | | |
|----|---------|----|-------------|------|
| 1. | what | IS | photosynthe | SIS! |

2. How is photosynthesis related to fluorescence?

3. How can fluorescence be used on an ecosystem scale and for what purpose?



Purpose: Crushing beetroot gives a bright purple liquid and crushing beet greens gives a bright green liquid, as you would expect from the appearance of these parts of the plant. However, the fluorescence of the liquids can be quite surprising due to the absorption and emission spectrum of chlorophyll. The change in appearance of the liquids in the fluorescence viewer will help illuminate the difference between absorptive color and fluorescence..

Protocol

- 1. Beets can be purchased in the grocery store with both leaves (greens) and attached roots. Cut up a small amount of beetroot with a knife or razor blade (\sim 0.1 gram) and add to a microcentrifuge tube. If you are using microcentrifuge tubes with graduations, you can fill the tube to the 100 μ L mark. You may want to use gloves when handling beetroots because they can stain your hands.
- 2. Add 500 μ L of isopropanol to the tube and use a pipette tip to crush the beetroot.
- 3. Tear up a small amount of beet greens (\sim 0.1 gram) with your hands and add to a microcentrifuge tube. If you are using a microcentrifuge tube with graduations, you can fill it up to the 200 μ L mark. Do not stuff the leaves into the bottom of the tube.
- 4. Add 500 μ L of isopropanol to the tube and use a pipette tip to crush the beet greens.
- 5. Set both tubes in a rack and wait for the large particles to settle.
- 6. Carefully pipette the supernatant from each sample into a clean tube being careful not to disturb the particles at the bottom. Try to recover at least 300 μ L Observe the color of the two tubes and record your observations in room light in the table below.
- 7. Place the tubes with the liquid in the DNA fluorescence viewer and place the photo hood on top. Take a picture with your cell phone and record your observations in the fluorescence viewer in the table below.

Observations

| Condition | Beetroot | Beet greens |
|-------------|----------|-------------|
| Room light | | |
| The Winston | | |



Questions

| 1. Compare the appearance of the liquid extracted from the beet roots under normal light and in the fluorescence viewer. What accounts for the color change? |
|---|
| 2. Compare the appearance of the liquid extracted from the beet greens under normal light and in the fluorescence viewer. How does the appearance of the liquid change when the photo hood is placed on top? Why? |
| 3. Put a small piece of beet greens in the fluorescence viewer near the light source. What do you observe? How do you explain the difference in appearance between the beet greens and the liquid extracted from the beet greens? |
| <u>Extension:</u> Repeat the protocol above with water instead of isopropanol. What differences do you observe when the tubes are placed in the fluorescence viewer? |

