# Thanks for the Support!



Principle of Plant Biology #6

Cell walls provide structural support for the plant and also provide fibers and building materials for humans, insects, birds and many other organisms.

Plant cell walls provide structure and support, even after a cell dies. Consider a tree – most of the cells in the trunk are no longer living, but the cell walls they leave behind are still functioning to support the tree. Materials made from plant cell walls are incredibly important in our lives. Look around the room – can you find ten things that are made from plant cell walls? These will include anything made out of paper, cardboard, cotton, or wood. Imagine if they all disappeared right now!

Cell walls change as a cell's needs change. Young, dividing cells have thin, flexible walls, whereas older cells may have extremely thick, rigid inflexible walls. By modifying the properties of their walls, plant cells optimize their functions. One type of plant cell with thick cell walls is called a fiber. Fibers provide structure and help plants carry water. Most wood and paper products are loaded with fibers from pine trees or other plants. Cell walls are chemically very strong. We use the strength and durability of the walls when we build shelters from wood, or when we make cotton clothing or paper. We use the stored chemical energy when we build wood fires – burning wood releases the stored energy as heat and light. We can also convert the energy in cell walls into liquid fuels like ethanol that can be used in place of gasoline to drive cars. This kind of renewable energy, or biofuel, may become very important as the cost of oil increases.

Plant cell walls are important in the diets of both people and animals. Our bodies are unable to release the chemical energy stored in cell walls, so most of it passes through our bodies undigested. This "roughage" or "fiber" helps maintain a healthy digestive tract. Some animals, like cows and termites, have microorganisms that live in their guts that help them to digest the cell walls and extract energy from them.



#### **Real-world Connection:**

Wood and plants are renewable resources that are continually replenished as they are used. Most houses are built from wooden frames. Many animals, such as birds, also rely on wood for their homes.

# Activity 1: Fibers in paper and cotton products

#### **Procedure:**

- 1. Tear or cut small pieces of different kinds of paper and cotton materials. Place several onto a microscope slide.
- 2. Place a drop of the blue-colored dye (Toluidine Blue – pronounced tall-you-ideen) onto each piece. Toluidine Blue stains most plant cell walls reddish purple, and stains those which contain lignin a bluish green. Lignin adds thickness and stiffness to cell walls.
- 3. After about a minute, blot away the excess stain, rinse the materials by dropping water onto them and blotting the excess water away. Place a few more drops of water onto the slide, then place a coverslip onto the slide.
- 4. Place the slide under a microscope and examine the materials using the lowest power lens.

#### **Observations:**

Sketch what you see below or on a separate sheet of paper. Be sure to label each drawing with the type of material it represents. Which of the products have the most air space between the fibers? Which is the most densely packed with fibers?

#### **Activity 2: Fibers in plants**

#### **Procedure:**

Your teacher will give you some plant materials and instructions on how to section them. "Sectioning" means cutting a paper-thin piece of something. Once you have made (or been given) sections, stain them with Toluidine Blue and examine them under a microscope as you did in Activity 1.

#### **Observations:**

Sketch what you see below or on a separate sheet of paper. Where in the plant sections are fibers (or other cells with thick cell walls) located? Do these fibers look like the ones in the paper and cotton products?

#### **Student-Designed Experiments**

Using the methods you learned in the demonstration above and the "Guide for Student Experimentation" below, design and carry out your own inquiry. Questions you might consider include the relationship between fiber or cell wall density and stiffness, the location and abundance of thick cell walls and fibers in various plant tissues, etc.

# **Guide for Student Experimentation**

### **Guidelines for Achieving Great Experiments**

- 1. Ask a very specific, testable question.
- 2. Test a control for comparison (a group that does not receive the experimental treatment).
- 3. Use a sample size large enough to allow firm conclusions.
- 4. To understand a whole population, obtain a random sample of that population to avoid bias.
- 5. Replicate each part of the experiment (at least 3 times).
- 6. Hold all variables constant between trials except the variable being tested.
- 7. Collect quantitative data whenever possible.
- 8. Measure using metric units.
- 9. Gather data carefully and accurately.
- 10. Be objective and honest.

#### **Introduction**

**Question:** 

Hypothesis:

#### **Materials and Methods**

**Independent variable:** 

**Dependent variable:** 

**Experimental constants:** 

**Control:** 

**Protocol:** 

# **Results**

Data collected:

**Other observations:** 

**Graph(s):** 

**Discussion** 

Interpretation of data:

**Conclusions:** 

# **Teacher's Guide to "Thanks for the Support!"**

# Links to National Science Education Standards

Grades 5-8:

- Abilities necessary to do scientific inquiry
- Understandings about scientific inquiry
- Structure and function in living systems

• Populations, resources, and environments

Grades 9-12:

- Abilities necessary to do scientific inquiry
- Understandings about scientific inquiry
- Form and function
- Natural resources

## **Materials**

- Toluidine Blue stain (available as solid or aqueous solution for \$10–30 for an ample supply).
- Droppers or dropper bottles for the Toluidine Blue stain (one per four students is ideal; one per class is manageable)
- Water with droppers in dropper bottles
- Compound microscopes, slides, and cover slips
- Single- or double-edged razor blades (single-edged are available from science supply companies; double-edged are available from grocery stores and make much thinner sections, but are more expensive and could be more dangerous)
- Products made from plant fibers including paper, facial tissues, cotton balls, cotton fabrics, etc.
- Plants to section (from grocery stores, gardens, school grounds, etc.) including celery, carrot, ginger root, potato and

any other plant materials the teacher or students want to examine.

# **Teaching hints**

**1. Preparing Toluidine Blue solution:** An aqueous solution can be purchased, but the powder form is cheaper. To prepare a 1 % solution, dissolve 1 g in 100 ml. Provide the students with glass or plastic droppers.

**2.** Set-up: Provide each team (of 4 students) with plant materials, 4 microscope slides, a squirt bottle of water (or beaker with water and a dropper), and a single-edged razor blade or a double-edged razor blade that has had one cutting surface covered with tape. Cover the tables with newspaper or other absorbent material to make clean-up easier. Gloves are useful when handling the Toluidine-Blue stains, but not necessary if the students are supervised and instructed to avoid contact with the stains. The stains are relatively non-hazardous but Toluidine-Blue can stain skin, nails and clothing.

**3. Safety:** If giving students razor blades is a concern in your classroom, then have a designated station for all cutting to take place.

**4. Interpreting Toluidine Blue staining of plant cell walls:** Toluidine Blue stains most plant cell walls reddish purple, and stains lignified plant secondary walls bluish green.



Toluidine Blue stained slice of a buttercup stem.

Tissue Element	Color Developed by
	Toluidine Blue O
Tracheary elements	Green or bluish green
(lignified walls)	
Lignified sclerenchyma	Blue-green, but
	occasionally green
Collenchyma	Reddish purple
Parenchyma	Reddish purple
Sieve tubes and	Red
companion cells	
Callose, starch	Unstained

Table 1. Plant cell staining by Toluidine Blue O (from O'Brien et al., 1964)

5. Tips for free-hand sectioning of plant materials: The teacher should experiment with making free-hand sections ahead of time, and demonstrate good technique before handing out the razor blades to the students. For preparing free-hand sections, double-edged razor blades are vastly superior to single-edged blades, because they are much thinner and flatter. However, they are also more dangerous. If you choose to use double-edged razor blades, you should cover one of the cutting edges with tape to prevent accidents.

Cutting is easier when both the blade and the plant material are wet. Set out cups of water to dip the blade between each slice. Don't anticipate that you will be able to slice all the way through your target tissue – instead, focus on getting very thin partial slices.

Hold the plant material in one hand. Hold the blade in your other hand and rest it on top of the plant material. Draw the blade along the surface of the tissue nearly parallel with the top surface (i.e. imagine you are making an invisibly thin slice). Transfer each slide to a dish of water or directly to a microscope slide – keep only the thinnest sections. Place a drop of water on the sections as you work to keep them from drying out.

Make both longitudinal (along the axis of growth) and transverse (cross) sections; the ginger root looks particularly nice in longitudinal section.

#### 6. Successful student-designed experiments:

- Emphasize the "Guidelines for Achieving Great Experiments."
- Before students design experiments, tell them how much time they will have.
- Allow students to present their experiments and lead a short discussion about each one. Encourage other students to ask questions.

#### References

- O'Brien, T. P., N. Feder, and M. E. McCully. 1964. Polychromatic staining of Plant cell walls by toluidine blue O. Protoplasma, 59: 367-373.
- Yeung, E. 1998. A beginner's guide to the study of plant structure. Pages 125-142, in Tested studies for laboratory teaching, Volume 19 (S. J. Karcher, Editor). Proceedings of the 19th Workshop/ Conference of the Association for Biology Laboratory Education (ABLE), 365 pages. http://www.ableweb.org/volumes/vol-19/9-yeung.pdf.

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