

## BioFuels: Cellulose Lab Investigation Sheet

**Driving Question:** How is biomass processed to become a biofuel?

By completing this activity you will:

1. Investigate how to prepare a biofuel source for conversion to a combustible product.
2. Understand that the refining process for biofuels requires an enzymatic reaction.

Products from plants can be used as a source for **biofuels**. All plant cells have cell walls that contain **cellulose**. Wood products such as paper, rayon, or cellophane retain wood's cellular structure. Cellulose is a polysaccharide, otherwise known as a complex sugar. Humans cannot digest cellulose. Cellulose is what we call dietary fiber.

Some animals and insects can digest cellulose and release the plant's sugars. These sugars provide energy. These animals may have special symbiotic microorganisms and/or digestive **enzymes** that breakdown the cell walls. **Enzymes** are proteins that help specific reactions. They can help make the starting substance turn into different products. **Cellulase** is one such enzyme. When plant fibers are exposed to cellulase, it allows the sugars in plants to be released. These sugars then **ferment** becoming **cellulosic ethanol**, a form of alcohol that can be used as an **alternative fuel source**. Presently cellulosic ethanol is used as an additive in gasoline.



### Step 1: Conducting the Experiment

1. Mark test tubes 1-4 on the white label space with wax pencils.
2. Store test tubes in test tube racks to keep upright.
3. Measure and place marks with wax pencils at **3 cm** and **6 cm** from the bottom of the test tube.
4. **Put goggles on.**
5. Fill test **tube 1** with **overnight cellulase/ pulp solution** to **6 cm** mark.
6. Fill test **tubes 2, 3, and 4** with **pure pulp** solution to the **3 cm** mark.
  - a. Fill **tube 2** to the **6 cm** mark with **5% cellulase solution**.
  - b. Fill **tube 3** to the **6 cm** mark with **rubbing alcohol**.
  - c. **Tube 4** is the **control tube**. Fill to the **6 cm** mark with water.
7. Complete the **first column** in the **“What is in your test tube?”** data table. Describe the solution you placed in the test tube. What was added to the paper pulp?
8. Predict which solution(s) will show that sugars were released from the cellulose after heating. Write your predictions in the **Prediction Section** on the next page.
9. Add **10 drops** of **Benedict's reagent** to each test tube.
10. Fill out **“What was the color of the solution before heating?”** on your data chart.
11. Carefully heat the test tubes by placing tubes in a hot water bath for **five minutes**.
12. Remove test tubes from the hot water bath using test tube clamps. Return tubes to test tube rack to cool.
13. Complete the data table.

- a. Fill out **“What was the color of the solution after heating?”** on your data chart. Note any color change in the test tubes.
- b. If sugar is present the solution, it will change color to green, yellow, orange, red, brick-red, or brown. The color is determined by the sugar present.

14. After the test tubes cool, empty liquids into the sink. Empty any solids into the trash.

15. Wash, rinse, and dry test tubes.



**Prediction Section**

- 1. Which solution do you think will release the sugars from the cellulose pulp? **Why?**



**Data Collection**

Complete the table below:

**NOTE:** A color change indicates the presence of a reducing sugar.

**What is in your test tube? Data Table**

Test Tube	Describe the paper pulp solution. [Was cellulase used in this sample? If so how long was the paper pulp in contact with the cellulase?]	What was the color of the solution before heating?	What was the color of the solution after heating?	Was sugar present in the solution?
1				
2				
3				
4				



## Step 2: Analysis and Conclusions

1. Which solutions showed a change in color?
2. What did the color change indicate?
3. What was added to the pulp solution that allowed the sugars to be released? Justify your answer with evidence from the laboratory.
4. Look at your results. Did the time the paper pulp was exposed to the cellulase, water, or alcohol appear to make a difference in the presence of detectible sugars?
5. Explain **why** or **why not** you thought the time the paper pulp was exposed to the cellulase, water, or alcohol made a difference.
6. Based on your observations, how do you think sugars are used to create biofuels?

