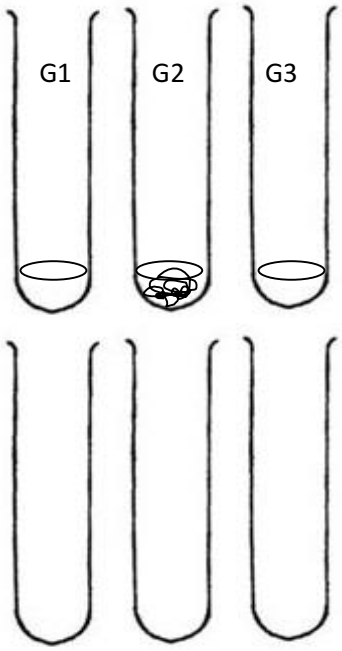
PART A – Investigating "Accellerase"

Your task is to determine whether the "Accellerase" is a carbohydrate, protein, or lipid. (We already know it is NOT a nucleic acid.)

There are chemicals (we'll call them "indicators") that change color in the presence of certain biological molecules. You will work as a group of four to complete the following four tests using these "indicators". Each person can complete one test and you can share the data with one another.

For each test, you will need 3 tubes. One will be a negative control (so that you know what a negative result looks like), one will be a positive control (so that you know what a positive result looks like), and one will be the "Accellerase" test result itself.

Sugar test--- Benedicts
Protein test—Biurets
Starch test—Iodine
Lipid test—Sudan IV



Negative test (G3 resembles negative control)

Positive test (G3 resembles positive control)

<u>Sugar test using BENEDICT'S Solution</u>

1. Label Three tubes G1, G2,and G3
2. Place 20 drops of distilled water in tube G1

3. Place 20 drops of the glucose solution in tube G2
4. Place 5 drops of the "Accellerase" in tube G3 and dilute with 15 drops of distilled water.
5. Add 4 drops **Benedicts** to tubes 1, 2, and 3. (read the label carefully—don't confuse it with Biurets!)
6. Place in hot water bath for 5 minutes. THIS IS THE ONLY INDICATOR TREATMENT THAT REQUIRES HEATING IN A HOT WATER BATH
7. Record the color of each tube in the data table.

Starch test using IODINE.

1. Acquire a set of 3 clean tubes. Label them S1, S2, and S3
2. Place 20 drops of distilled water in tube S1
3. Place 20 drops of starch solution in tube S2
4. Place 5 drops of the "Accellerase" in tube S3 and dilute with 15 drops of distilled water.
5. Add 4 drops Iodine to tubes S1,S 2, and S3
6. Record the color of each tube in the data table.

Protein Test using BIURETS

1. Acquire a set of 3 clean tubes. Label them P1, P2, and P3
2. Place 20 drops of distilled water in tube P1
3. Place 20 drops of albumin solution in tube P2
4. Place 5 drops of the "Accellerase" in tube P3 and dilute with 15 drops of distilled water.
5. Add 4 drops Biuret's solution (read the label carefully—don't confuse it with Benedicts!) to tubes P1, P 2, and P3
6. Record the color of each tube in the data table.

Lipid Test using SUDAN IV

1. Acquire a set of 3 clean tubes. Label them L1, L2, and L3
2. Place 20 drops of distilled water in tube L1
3. Place 15 drops of distilled water and 5 drops of vegetable oil in tube L2
4. Place 5 drops of the "Accellerase" in tube L3, and dilute with 15 drops of distilled water to dilute
5. Add a small amount of Sudan IV powder (the amount you can "scoop" with a toothpick) to tubes P1, P 2, and P3. Swirl gently.
6. Record the color of each tube in the data table

Name _____

Section _____

PART A – Investigating "Accellerase"

	Sugar Test	Starch Test	Protein Test	Lipid Test
Name of Indicator				
Heat required?				
Materials in negative control				
Color of negative control				
Materials in positive control				
Color of positive control				
Color of material in tube "3" Experimental tube				

BE SURE YOU SHOW YOUR TEACHER THE RESULTS OF YOUR TABLE'S TESTS BEFORE YOU CLEAN UP!!!

- Explain why it makes sense that biofuels will be based on carbohydrates or lipids.

_____
_____
_____
_____.

- Explain the relationship between cellulose and glucose, using the terms monomer and polymer in your description.

_____
_____
_____
_____
_____
_____

- Once an inexpensive source of sugar is obtained, what is the next step, and what kind of organism is required?

_____
_____
_____
_____
_____
_____.

- What kind of biological molecule is "Accellerase"?
  _____

- What is the purpose of a control group?

_____
_____
_____
_____
_____
_____

- Why is a positive control and a negative control needed to determine what kind of biomolecule "Accellerase" is?

_____

_____
_____
_____
_____

Part B - Using Switchgrass and "Accellerase" to obtain sugar.

Now that you know what kind of molecule "Accellerase" is, we will use it to actually break down cellulose polymers into sugar monomers. Recall that the sugar will be used in a subsequent step to produce ethanol, which is a flammable liquid suitable for many automobile engines.

We won't be using pure cellulose, because purifying cellulose to produce ethanol would prevent it from being a cost-effective source of sugar. We will use dried plant material from a plant that grows readily in New York state→ switchgrass! You will work as a group to set up the treatments, and will collect data next week in lab.

It is not sufficient to know that "Accellerase" works. We want to optimize the production of sugar by this molecule, so we will conduct some preliminary experiments.

Factor A: pH.

A water molecule is made of two hydrogen atoms and one oxygen molecule. Some water molecules break down into H+ and OH- ions. Pure water is considered neutral and has a pH of 7 because it has an equal number of H+ and OH- ions.

If there are more H+ ions in the solution than OH-, the solution will be acidic. Acidic solutions have a pH less than 7. pH is on the logarithmic scale, which means that a pH of 6 will be 10 times more acidic than a pH of 7, and that a pH of 5 is 100 times more acidic than a pH of 7.

If there are more OH- ions in the solution than H+ ions, the solution will be basic. Basic solutions have a pH more than 7. Because pH is on the logarithmic scale, a pH of 8 is 10 times more basic than a pH of 7, and that a pH of 9 is 100 times more acidic than a pH of 7.

The H+ ions and OH- are charged and often interact with other molecules. Many kinds of molecules are permanently damaged by strong acids and strong bases. You will investigate whether "Accelerase" is affected by the pH of the solution.

******EVERY GROUP WILL EVALUATE THE EFFECT OF pH ON ACCELLERASE.******

Factor B: Switchgrass pre-processing.

One third of the class will perform the pH study using dried switchgrass

One third of the class will perform the pH study using pelletized switchgrass

One third of the class will perform the pH study using powdered switchgrass.

Data regarding switchgrass pre-processing will be compiled later, and you will be expected to draw conclusions based on the data.

Procedure, Day 1

1) Label tubes 1-8, including your name and the date.
2) Pour 15 ml of distilled water into tubes 1-8.
3) Add 0.5g of switchgrass material (dried or pelletized or powdered) to tubes 1-8.
4) Create a solution of pH 3 to tubes 1 and 5. (~40 drops of HCl). Use a pH strip to determine pH and adjust until the solution reaches the appropriate pH.
5) Create a solution of pH 5 to tubes 1 and 5. (~15 drops of HCl). Use a pH strip to determine pH and adjust until the solution reaches the appropriate pH.
6) DO NOT add HCl or NaOH to tube 3 or tube 7. Use a pH strip to determine pH and record.
7) Create a solution of pH 9 to tubes 4 and 8. (~10 drops of NaOH) Use a pH strip to determine pH and adjust until the solution reaches the appropriate pH.

8) Add 0.5 ml of "Accelerase" to tubes 1-4. **DO NOT add "Accellerase" to tubes 5-8.**

9) Wrap your tubes or test tube rack with a piece of tape that includes your group's name and date. Hand prepared tubes to your teacher, and wait to see how well "Accellerase" worked the next time the lab meets (1 day -1 week, as needed).

Hypothesis formation:

Do you think pH will have an effect on "Accellerase" effectiveness?
Yes or No

Do you expect to see a difference between the acid treatment and the base treatment?
Yes or No

Do you expect to see a difference between the whole switchgrass, pelletized switchgrass, and powdered switchgrass treatments?
Yes or NO

Which treatment do you think will result in the most sugar? _____

Explain your reasoning (why do you think you will see the results you predict) in three to four sentences.

Procedure, Day 2

1) Obtain your tubes from the previous lab

2) Read instructions on using the glucose meter and glucose strips.

3) Measure the glucose in each of your tubes, and record your group's data in the table below.

	pH of solution (day 1)	"Accellerase" in tube (yes or no)?	Glucose present in tube after treatment (day 2)
Tube 1			
Tube 2			
Tube 3			
Tube 4			
Tube 5			

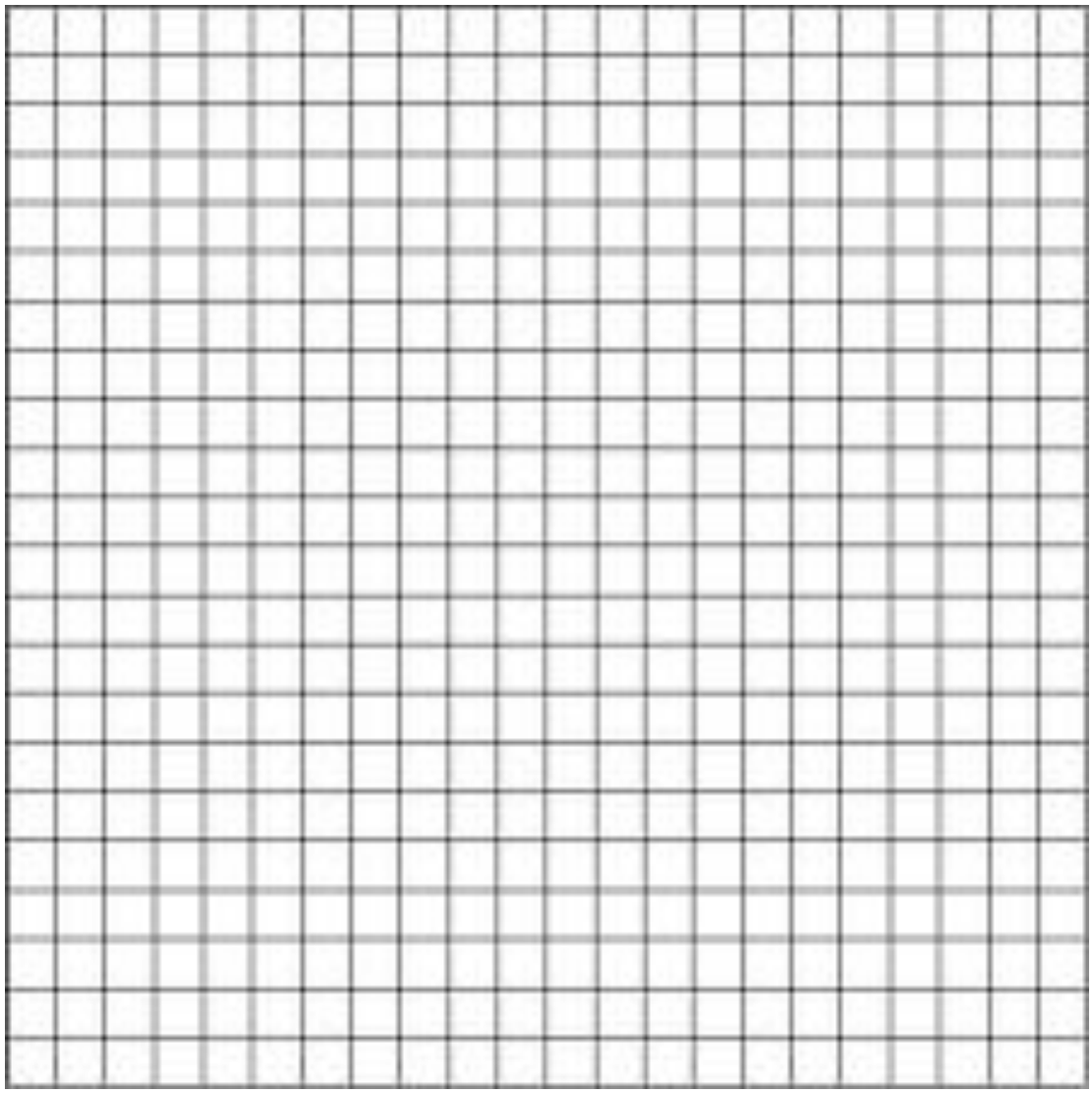
Tube 6			
Tube 7			
Tube 8			

On the following page, graph your data!

The pH should be on the x-axis (because it is the _____ variable, which is under the control of the scientist).

The amount of glucose should be on the y-axis (because it is the _____ variable, which the scientist measures at the end of the experiment)

Make two separate lines—one for the tubes with "Accellerase" and one for the tubes without "Accellerase". BE SURE TO LABEL THE LINES SO YOU CAN TELL WHICH IS WHICH!!!



Analysis of your group's data

Based on your results, did "Accellerase" improve the sugar produced from switchgrass? Explain.

Based on your results, what pH range that allowed the "Accellerase" to work best? Explain. (Scientists would conduct more experiments to determine the "optimum" pH for sugar production).

Average glucose levels in treated switchgrass material

	Cut	Pelletized	Powdered
Tube 1			
Tube 2			
Tube 3			
Tube 4			
Tube 5			
Tube 6			
Tube 7			
Tube 8			

Looking at the class data, did adding "Accellerase" to the tubes result in more sugar production for all groups?

Looking at the class data, which pH range seemed to produce the most sugar ("Accellerase" treatment)?

Looking at the class data, was there a pretreatment that resulted in more sugar production than the others? Which one? Try to provide an explanation for that result below.

You have good preliminary results for making sugar from switchgrass and "Accellerase". What conditions would you start with if you wanted to start your own company—one that would use "waste" cellulose from vegetable waste to produce sugars for ethanol production?