BIOFUELS AND BIOPRODUCTS (HS)

Biomass to sugars

Focus questions	Which feedstock will produce the most glucose for fermentation?					
Vocabulary	Polysaccharide, disaccharide, monosaccharide					

How much glucose is available in sweet corn and dent corn for ethanol production? You will investigate the glucose content in both sweet and dent corn over a 3 day period. During this process, you will compare data and analyze the treatment methods employed to extract glucose from the feedstocks. You will construct (and revise) an explanation for which feedstock provides the most glucose for fermentation over time. You will have an opportunity to continue to research ethanol production and propose future solutions to improve the efficiency of ethanol production.

Materials

- 1 test tube rack
- 10 test tubes or centrifuge tubes
- 10 mL graduated cylinder
- Marker
- Stir rod
- Glucose, ground up
- 2 types of corn, ground up (sweet, dent)
- Scale
- Amylase solution
- Glucoamylase solution
- Hot plate
- · 500 ml beaker that is half full of water
- .5 ml disposable pipette.
- · Hot gloves
- Safety goggles
- Access to water, glucose test strips or glucose monitor and test strips, enzyme complexes and a scale are needed.

Procedure

- 1. Work together in groups of 4 for the investigation.
- 2. Use your marker to label your tubes according to Data Table 1.
- 3. Using a graduated cylinder to measure (or the marks on the centrifuge tube), pour 10 ml of water from your beaker into each test tube. Then return the test tube to the rack.
- 4. Measure 1.0 g of glucose or ground corn into each of the appropriate labeled test tubes. Note that the enzyme only tube does not get any glucose or corn. When you are done, put the caps back on all 10 tubes.
- 5. Make sure your beaker is half full of water and place it on the hot plate until it begins to boil. Wear safety goggles and use caution around the hot plate!

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- 6. Loosen the caps on the test tubes labeled "heat." Carefully place the 3 test tubes labeled "heat" in the beaker of boiling water. Allow samples to sit in the gently boiling water for 10 minutes. Reminder: If it appears that water may begin to spill out, turn the heat down.
- 7. After 10 minutes, remove the tubes using a hot glove, and place back in the test tube rack.
- 8. Once the test tubes are cool enough to touch without the glove, carefully tighten the caps on the tubes and cool the heated tubes by running them under cool water or placing them in an ice water bath. Once samples have cooled to room temperature, move on to step 8.
- 9. Add .5 ml of Amylase and Glucoamylase solution to glucose/heat, glucose/ no-heat, sweet corn/heat, sweet corn/heat, dent corn/heat, dent corn/no-heat and enzyme only test tubes.
- 10. Cap and swirl to mix gently, then proceed to step 10.
- 11. To measure sugar, dip 1 glucose strip in a tube for 1–2 seconds. Be sure that the entire pad on the end of the strip is submerged in the solution. *
- 12. Remove the test strip from the sample and gently drag it across the lip of the test tube to remove any excess liquid. Then prop the test strip up on its side so the liquid runs off the strip instead of pooling on the test area, and let sit for 1 minute. Next, dip a new test strip for the next test tube and start timing 1 minute. While the other team members move on, be careful to keep track of which strip goes with which tube.
- 13. Compare test strips with the color chart on the bottle and record your results in the data table. Be sure to do this right after the initial 1 minute is over because the color change on the glucose test strip will be inaccurate after 2–3 minutes.
- 14. Repeat steps 10 through 12 until your team has measured and recorded sugar levels for the samples in all 10 test tubes. Alternatively, If a glucose monitor is present, place a test strip in the machine and turn it on, use a pipette to place a drop of solution on the end of the test strip at the prompt, read and record measurement at the prompt. Once you are done testing the glucose levels, place the caps back on the test tubes and loosely tighten.
- 15. Incubate at room temperature overnight, or, for optimal enzyme function, place in an incubator or water bath at 50–60°C overnight. Day 2 and 3
- 16. Measure sugar levels the same way you did in steps 10-13 above (Sugar Measurements) for all samples during your next 2 classes. Before taking sugar measurements, check that cap tubes are tight and gently swirl each tube for a few seconds to mix. Then measure and record the amount of sugar in all tubes using the glucose strip, or glucose monitor, as you did on Day 1. Place your data into the chart below.

Table 1: Data

Tube no.	Label	Glucose amount day 1: time 0	Glucose amount day 2: time hours	Glucose amount day 3: time hours		
1	Glucose / heat / date					
2	Glucose / no heat / date					
3	Glucose / no treatment / date					
4	Sweet corn / heat / date					
5	Sweet corn / no heat / date					
6	Sweet corn / no treatment / date					
7	Dent corn / heat / date					
8	Dent corn / no heat / date					
9	Dent corn / no treatment / date					
10	Enzyme only / date					

Graph the data from the chart above. Show each tube individually as a line in the graph below.

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Analyze your data.

Reflection

Reflect on the following questions while creating your explanation for which feedstock will provide the most glucose for fermentation over time and design a solution to use the most logical feedstock(s) in the United States.

- 1. Where does the glucose come from in sweet corn or dent corn?
- 2. How much glucose is present in each feedstock for ethanol production?
- 3. Which type(s) of pretreatment were most effective at liberating glucose? How does each pretreatment manipulate the feedstock?
- 4. What additional pretreatments for dent corn can be employed to improve the efficiency of ethanol production?
- 5. What other feedstocks can be used in ethanol production?
- 6. What possible alternative feedstock options are available that can help to make ethanol production more efficient and sustainable in the future?
- Construct and revise an explanation to determine which feedstock will provide the most glucose for fermentation over time and design a solution to use the most logical feedstock(s) in the United States.

Rubric for self-assessment

Skill	Yes	No	Unsure
I can explain the difference in glucose content between sweet corn and dent corn for ethanol production.			
I can use reasoning to connect the evidence of glucose availability in starch and construct an explanation to demonstrate why dent corn is used in commercial ethanol production.			