Gassy Microbes

SC Academic Standards: 4.L.5B; 5.L.4B; 6.L.4A; 6.L.4B; 7.L.3A; 7.L.3B; 7.EC.5B; H.B.2; H.B.3A; H.B.6.

NGSS DCI: 4-LS1.A; MS-LS1.A; HS-LS1.A

Science and Engineering Practices: S.1A.1; S.1A.2; S.1A.3; S.1A.4; S.1A.5; S.1A.6; S.1A.7

Crosscutting Concepts: Cause and Effect: Mechanism and Explanation; Energy and Matter; Stability and Change, Systems and Systems Models.

Focus Question(s): How do we measure the rate of cellular respiration in yeast? How does pH and temperature affect the rate of cellular respiration?

Conceptual Understanding: All organisms need energy to live and grow. Energy is obtained from food. The role an organism serves in an ecosystem can be described by the way in which it gets its energy. Energy is transferred within an ecosystem as organisms produce, consume, or decompose food. A healthy ecosystem is one in which a diversity of life forms are able to meet their needs in a relatively stable web of life.

Life is the quality that differentiates living things (organisms) from nonliving objects or those that were once living. All organisms are made up of cells, need food and water, a way to dispose of waste, and an environment in which they can live.

The Fungi Kingdom consists of organisms that do not make their own food (heterotrophs) but obtain their nutrition through external absorption. Fungi can be grouped by their growth habit or fruiting structure and respond to changes in the environmental stimuli similar to plants.

Cells transform energy that organisms need to perform essential life functions through a complex sequence of reactions in which chemical energy is transferred from one system of interacting molecules to another.

Background: Some microbes produce different types of gases as a byproduct of their metabolic processes (such as decomposition or digestion). The microbes in this Science Friday Video (http://www.sciencefriday.com/blogs/01/25/2010/gassy-microbes.html?audience=2) released an odorless and flammable gas called methane. The type of gas or gases released by a microbe depends on the species and their metabolic characteristics.

In this activity, students will conduct an experiment to observe a metabolic process, the breakdown of glucose sugar, by yeast cells. Students will vary conditions in the yeast's surrounding environment and observe the amount of gas that the yeast releases. Students will use the results to determine which environmental conditions are suitable for yeast growth.

Yeast is a single-celled organism that can only be seen through a microscope. We are able to see the yeast in this experiment because a packet of dry yeast sold in a grocery store holds billions of yeast cells (one gram holds about 25 billion cells). These yeast cells lie in a dormant state until they are activated by the warm water, and begin to break down the sugar for energy. As the yeast cells metabolize the sugar, they release carbon dioxide gas. The build up of carbon dioxide gas in the bottle causes the balloons to inflate.

Many scientists are interested in the effects of natural recycling that takes place in the Earth's biosphere. Recycling biodegradable material often requires the presence of moisture for the microorganisms (creatures responsible for the biological breakdown of trash) to be activated.

The yeast in this experiment consists of living organisms that break down (decompose) the substrate (sugar and water) and produce carbon dioxide gas. Although its hard to see a microorganisms without a microscope, you can see evidence (the carbon dioxide) of them "eating", which is how they break down food sources (in this case sugar) that provide energy for their tiny systems and help recycle materials.

Similar to any other living organism, yeast need to be in an environment that is suitable for survival. The yeast's ability to thrive in the bottles, or the artificial "environments" that the students have created, can be measured by how well each balloon inflates. A thriving community of yeast will produce more carbon dioxide gas. If less carbon dioxide is produced, the environment for yeast is less suitable.

Materials: **Part 1:** ***for each student (or group)*** get three 16 oz. clear plastic soda or water bottles, Active dry yeast or baker's yeast, 3 Sugar packets, 1 Plastic spoon, 3 Small plastic cups, Warm water, Large bowls, Ice, Vinegar, Permanent marker, 3 Balloons (ask about latex allergies!).   **For part 2** you will also need, per group: 5-6 packets of baker's yeast, 5-6 (reuseable) ziploc bags, a one cup measure, a one - tablespoon measure, a one-teaspoon measure, a large bowl, a thermometer, sugar, a ruler, ice water, hot water, and white vinegar.

Previous Knowledge: (biology): acids, and heat, can denature enzymes. When the yeast enzymes don't work because they are denatured, the yeasts can't break down sugar into CO_2 gas (enzymes are necessary in cell respiration, which is the process that breaks sugar down into CO_2, H_2O, and ATP energy – it occurs in the mitochondria of plants and animals and fungi like yeasts). Often there isn't enough oxygen present for the yeast to break sugar down aerobically, so yeast switch to anaerobic fermentation and break sugar down into CO_2 and alcohol and much less ATP than if oxygen were present. But, since CO_2 is still produced, we still see reactions in our plastic bottles.

Yeasts are fungi, and fungi (as well as many bacteria) are decomposers. Decomposers break down dead (usually) bodies and help recycle their nutrients / atoms back into the soil, air and water. Thus, they are vital links in the carbon cycle for example, taking the carbon in the carbohydrates of a body and converting it to gaseous carbon dioxide which is released into the air.

<u>Procedure</u>: Part 1.

1. Begin the lesson by having students watch the Science Friday Video, "Bubbling Methane; Melting Permafrost." http://sciencefriday.com/teacher-resources/01/25/2010/gassy-microbes.html
2. Explain to students that permafrost is permanently frozen ground found in cold regions such as Alaska. Ask students what could cause permafrost to melt. How can melting permafrost affect the environment?
3. Tell students that they are going to conduct an experiment to observe how a microbe interacts with its surrounding environment, and how varying conditions in the environment can affect its ability to thrive.
4. Hand out three plastic bottles to each student. Have students use the permanent marker to label each of the plastic bottles in the following manner: Bottle A – Sugar, Bottle B – Vinegar, Bottle C – Sugar/Ice
5. Distribute to each student three sugar packets and three plastic cups with small portions of the following ingredients separated into each cup: yeast, warm water, and vinegar. Ask students to explain what yeast is, and how it is used. Why do they think we are using yeast in an experiment about microbes?
6. In Bottle A, have students mix one spoonful of yeast, one packet of sugar and four spoonfuls of warm water. Gently swirl the ingredients inside the bottle until the contents have dissolved. Cover the top of the bottle with a balloon. Have students observe the contents inside the bottle. Why is it bubbly or foamy? Does it remind them of any footage from the Science Friday Video?
7. In Bottle B, have students mix one spoonful of yeast, one packet of sugar, three spoonfuls of warm water and one spoonful of vinegar. Gently swirl the ingredients inside the bottle until the contents have dissolved. Cover the top of the bottle with a balloon. What is happening inside the bottle?
8. In Bottle C, have students mix one spoonful of yeast, one packet of sugar, and four spoonfuls of warm water. Gently swirl the ingredients inside the bottle until the contents have dissolved. Cover the top of the bottle with a balloon. Place the bottle upright in the middle of a large bowl, and place as much ice as possible around the bottle.

9. Set the bottles aside for about 15 to 20 minutes. During this time, have students create a chart on a sheet of paper with three columns and two rows. Label the columns “Bottle A”, “Bottle B,” and “Bottle C.” Label the top row as “Predictions” and the bottom row as “Results”.

	Bottle A (yeast, water, sugar)	**Bottle B (yeast, water, sugar and vinegar (=acid))**	**Bottle C (yeast, water, sugar, and placed in ice!)**
Predictions			
Results			

Table 1. Predictions and results for bottle experiment with sugar / yeast

10. Have students write their predictions in the appropriate column and row for each bottle. Ask students to write explanations for their predictions on the chart. Based on their initial observations, what do they think will happen to the balloons? Which balloon do they think will inflate the most? After students have finished writing their predictions, have them discuss their predictions and explanations with the entire class.
11. After 20 minutes, have students observe their bottles. What has happened to the balloons? Why have some balloons inflated more than others? Have students write the results in the bottom row of their chart. Compare and contrast their results with each other, and with the students’ predictions.

Part 2: Do you want data? Try it this way, using Ziploc bags instead of a balloon / bottle (we will measure, with a ruler, how much the bags inflate):

<u>**Materials**</u>: *per group:* baker’s yeast in packets, Ziploc bags, a teaspoon measure, one cup measure, sugar, a large bowl, 1 thermometer, 1 ruler, lab notebook and pencil. (optional: vinegar)

1. Pour one packet of activated dry yeast into each Ziploc bag.

2. Add 1 teaspoon of sugar to one of the bags and label the bag 1 tsp.

3. Add 2 teaspoon of sugar to another bag and label it as 2 tsp.

4. Add 4 teaspoon of sugar to the fourth bag and label it as 4 tsp.

5. Mark 0 on the outside of the last bag and do not add any sugar to it.

6. Pour warm water into the large bowl so it is about 2/3 full. Check the temperature of the water with the thermometer. The water should be about 46^{o}C (115^{o}F). Add hot or cold water to bring the water to this temperature.

7. Use the measuring cup to dip 1 cup of warm water from the bowl into each of the bags. Gently squeeze each bag between your fingers to mix the contents thoroughly. Make sure that there are no dry pockets of yeast or sugar in the bags.

8. Squeeze most of the air out of the bags and seal them. Set the bags in the bowl of warm water in a warm place so it will not cool down rapidly.

9. Every 5 minutes for 30 minutes: Take the bag marked 0 out of the water, dry it, and place it on a flat table. Put the cardboard or notebook on top of the bag, holding the tablet level. Use the ruler to measure the distance from the table to the bottom of the cardboard.

10. Record your measurements.

11. Repeat #9 with the remaining bags.

12. Calculate the approximate volume of carbon dioxide in each bag: measure the length of the bag (a); measure the width of the bag (b); distance from table to cardboard (c). Multiply a x b x c to get the volume in each bag.

Time (min)	**0 tsp sugar**	**1 tsp sugar**	**2 tsp sugar**	**4 tsp sugar**
0				
5				
10				
15				
20				
25				
30				

Table 1. Volume of carbon dioxide in bag

13. Repeat experiment varying first pH (by adding vinegar) and then by varying the temperature of the water. Try a bag with one cup water, a second bag with (one cup – 1 tbs water + 1 tbs white vinegar), a third bag with (one cup – 2 tbs water + 2 tbs vinegar) and so forth, then try a bag with 1 cup of ice water a second bag with 1 cup of room temperature water, a third bag with 40 degree water and a fourth bag with 80 degree water. Remember, you can only test one variable at a time (don't change both temperature and pH) - and use these experiments to reinforce the concepts of replication (class data) and controlled variables, and a control experiment.

Extensions: If you can combine the class data then you can take an average of those individual data (your replicates). It is always good to stress replication in science. Also stress the difference between a control experiment and a controlled variable. The **control** is your comparison group – it is how you can tell if your independent variable is causing your data to do something different than normal. Here, it is the 0 tsp sugar group. **Controlled** variables are the all the variables except your independent variable – the variables that should be the same in both control and experimental treatments.

Reflection Questions:

- **How does the acidic environment created in this experiment compare to the effects of acid rain in our environment?** (acid rain is rain with a pH that is lower than 7. Some areas in the US experience rain with a pH in the 4-5 range. White vinegar is about 2.4).

- **How would we create an experiment to simulate the effects of climate change in our environment? What variable would we have to change?** (temperature!).

- **Besides carbon dioxide and methane, what are other types of gases or by-products that may be released by microbes?** (NO = nitrous oxide – produced by soil bacteria)

- **Why did some balloons not inflate, or not inflate as much as others?** (The balloon for Bottle B will not inflate, because vinegar is a type of acid. An overly acidic environment can hinder the growth of yeast. Yeast tends to thrive in warm environments. By surrounding Bottle C with ice, we have created an environment that is too cold to sustain yeast growth. Bottle A should have the most inflated balloon of all three because it contains the optimal conditions for the growth of yeast -- or, in the plastic bag experiment The bags contain various amounts of carbon dioxide because more decomposition took place in the bags with larger amounts of sugar; the yeast in the bag marked 0 had no sugar, so no carbon dioxide was produced).

- **Explain the process of how yeast helps bread dough to rise.** (yeast breaks down the sugar and produces CO_2 which is a gas and causes the dough to rise. Without plentiful oxygen, yeast break down sugar in to carbon dioxide and alcohol, which is baked off in the oven (yeast also ferment grapes into wine, but in this case the alcohol isn't baked off, it remains in the mixture giving wine its alcoholic content)).

<u>Models and Explanations</u>: This experiment is cellular respiration, where yeast cells are breaking down sugar into CO_2, H_2O, and ATP. **A student who demonstrates understanding** of these concepts can explain what the carbon source is for the CO_2 gas that is produced, and can explain why CO_2 is not produced in some of the plastic bottles. This student can also link yeasts, as fungi, to the carbon cycle as major decomposers who convert the organic carbon (carbohydrates) bound up in living bodies back into inorganic carbon (carbon dioxide gas) which is released into the atmosphere. From there, plants uptake the CO_2 and turn it back into sugar, completing the cycle.

(**Teachers**: yeast are often facultative anaerobes – and even though there is oxygen in the baggies the yeast are pretty bad at getting the oxygen out of solution. Most likely, instead of aerobic respiration (oxygen + glucose $\rightarrow$ CO_2 + H_2O + ATP) we are seeing anaerobic respiration (glucose $\rightarrow$ CO_2 + ethanol + ATP). Either way, CO2 is produced and thus we know glucose is being broken down.

<u>Bibliography:</u>

Breathing Yeasties. Retrieved August 12, 2014 from http://www.howtosmile.org/record/529.

Sciencefriday.com (2010). Gassy Microbes. Retrieved September 7, 2014 from http://sciencefriday.com/teacher-resources/01/25/2010/gassy-microbes.html

Student Worksheet

In this activity, students will conduct an experiment to observe the metabolic process of yeast by using household ingredients. Students will vary conditions in the yeast's surrounding environment and observe the amount of gas that the yeast releases. Students will use the results to determine which environmental conditions are suitable for yeast growth.

	Bottle A (yeast, water, sugar)	Bottle B (yeast, water, sugar and vinegar (=acid))	Bottle C (yeast, water, sugar, and placed in ice!)
Predictions			
Results			

Table 1. Predictions and results for bottle experiment with sugar / yeast

Time (min)	0 tsp sugar	1 tsp sugar	2 tsp sugar	4 tsp sugar
0				
5				
10				
15				
20				
25				
30				

Table 2. Volume of carbon dioxide in bag

Time (min)	**0 tsp vinegar**	**1 tsp vinegar**	**2 tsp vinegar**	**4 tsp vinegar**
0				
5				
10				
15				
20				
25				
30				

Table 3. Volume of carbon dioxide in bag (acid conditions: vinegar added)

Time (min)	**Ice water (0^{oC})**	**Room temperature water ($25^{o}C$)**	**Warm water ($40\ ^{o}C$)**	**Hot water ($80\ ^{o}C$)**
0				
5				
10				
15				
20				
25				
30				

Table 4. Volume of carbon dioxide in bag (temperature experiment)

Please graph your results for tables 2-4.