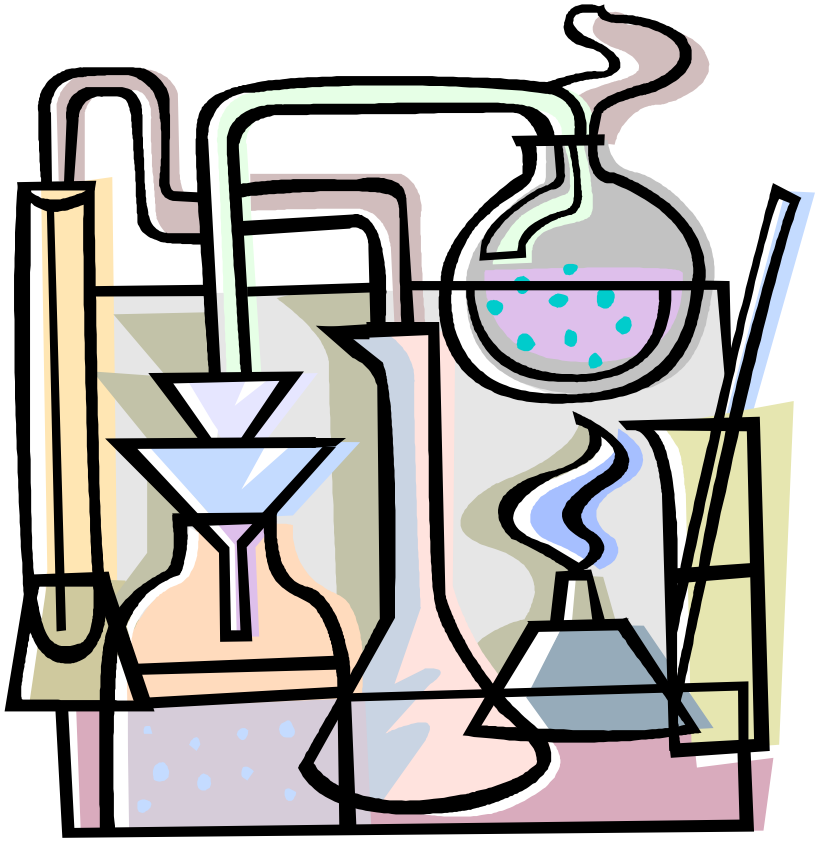
Discovering Enzymes



Support for Cornell Center for Materials Research is provided through NSF Grant DMR-0079992

Discovering Enzymes

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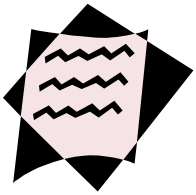
Abstract



Students use hydrogen peroxide to view reactions between enzymes and proteins and thank about the results.

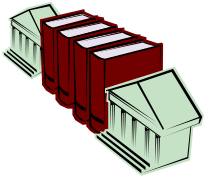
Equipment

1. Hydrogen peroxide
2. Pippettes
3. Test tubes
4. Gloves
5. Safety goggles
6. Potatoes
7. Eggs (egg whites)
8. Carrots
9. Dirt
10. Leaves
11. Wood
12. Rocks

Grade Level

This activity is suitable for Elementary and Middle School Students.

State Standards Met



Standard 1 – Analysis, Inquiry, and Design
Standard 4 – Physical Setting and Living Environment
Standard 7 – Application of Math, Science and Technology

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Enzymes – Activity Outline

Introduction - Put a small amount (about 1-inch high) of hydrogen peroxide into a test tube. Cut a small sliver of fresh potato and drop it into the hydrogen peroxide. Bubbles will start to form around the potato sliver. What's going on?

There are lots of questions you could ask about this reaction but this activity addresses two chief questions.

Activity One - What things make bubbles when immersed in hydrogen peroxide?

As a group, design experiments to test which things (milk, carrots, earth, leaves, wood, hair, spit, rocks, etc.) make bubbles in hydrogen peroxide. Think about how to make the comparison as accurate as possible. Predict (or guess) what you think will happen? Write down a description of your experiments, predict (or guess) the results you expect, carry out the experiments and summarize the results.

Activity Two - Can we speed up, slow down or stop the reaction of hydrogen peroxide and potato juice?

We will blend and separate potato juice. Mixing potato juice and hydrogen peroxide makes foam filled with bubbles. As a group, design a protocol to test the effect of different chemicals and conditions on the reaction. Don't forget to do a proper "control" experiment.

Summarize the results of all the tests to show the effect of chemicals and conditions on the reaction.

Conclusion -

To conclude, we'll do one last test of the stability/fragility of proteins. Shake potato juice with acetone (best known as nail polish remover) and test for activity: no activity. The potato juice doesn't change appearance when we add acetone because the concentration of the active protein, catalase, is very small. Try the same thing on egg white that contains a far greater concentration of proteins (in particular albumin). This time the egg white turns into a white hard solid just like cooked egg white. Acetone has completely changed the structure of the proteins in egg white. Bad conditions destroy the complex structure of proteins. When the proteins in egg white lose their structure, they turn white and gel together. When the catalase in potato juice loses its structure, it can no longer break down hydrogen peroxide.

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Enzymes – Notes for Instructors

Safety Information: The adult should also insist that hydrogen peroxide is a dangerous chemical: the children are then invited to protect their eyes with safety glasses and to avoid contact with the skin (and with their clothes, as far as possible, some colors could fade away), for example by wearing gloves, using pipettes to count hydrogen peroxide solution drops, and closing the bottles as soon as they are finished.

If a student spills chemicals on themselves, wash away the chemical with excess water.

Tips for the Activity: The aim of this activity is to play with proteins and discover their properties such as catalytic activity, structural features and stability/fragility. We will use proteins from very common sources:

- potato juice, in which we are interested in catalase, an enzyme which catalyses the dismutation (break-down) of hydrogen peroxide,
- egg white, in which we are especially interested in albumin, the main component, which has a structural role in the egg white gel,
- anything we can find in the room.

Fresh potato shows an interesting chemical activity. When dipped in a solution of hydrogen peroxide, it triggers bubbling of oxygen. This activity is due to a special protein produced by the potato to protect itself against oxidative stress. Oxidative stress is very common on our planet because of our oxygen rich atmosphere. Children could be invited to tell what they know about oxidation and give practical examples: they may know, for example, that iron is oxidized into rust by oxygen from the air, a process accelerated by water and salt. Or they may know that the skin is sensitive to oxidative agents called "free radicals" for which cosmetic manufacturers design special "anti-age" creams (often containing vitamin C as the anti-oxidizer). Or they may know that UV light shining on oxygen turns it into an even stronger oxidant, ozone (O3), which is in the ozone layer or in copiers, and that ozone is dangerous (everybody can recognize the smell of ozone because of copiers). The enzyme in potato is called catalase. An enzyme makes a reaction happen faster. If you let hydrogen peroxide sit in a container for long enough (months at room temperature) bubbles of oxygen would be released. The catalase in potato juice breaks the hydrogen peroxide down much, much faster.

What is nice with this activity is that the children are free to build their own experiment and to discover by themselves, so that you should not tell in advance what the result will be (very often, anyway, we would not know…).

For activity one, students should feel free to try many different materials. Adults should guide students to build a protocol for testing each substance in a safe and accurate way. Students should write a short description of the test, predict (or guess) the result they expect (and why, if possible), carry out the experiment and record the result. For example, a test of grass, salt and potato might go like,

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Example: Activity of grass and salt

Test Tube One: Three blades of grass + 10 drops of hydrogen peroxide solution.

Test Tube Two: Volume of salt (approximately equal to volume of grass) + 10 drops of hydrogen peroxide solution.

Test Tube Three: Slice of potato (same volume as salt and grass) + 10 drops of hydrogen peroxide.

Measurement: Watch each test tube carefully for 2-3 minutes to see how many bubbles form.

Prediction: The salt won't do anything. Salt isn't alive. Grass will work just like potatoes.

Result: ?????

We included the potato so we could compare the activity of each substance to potato, as we know that works!

In general, catalase and other peroxidases are present in all plants and animals living under aerobic conditions for detoxification (although their exact roles are not well understood, in part because the role of oxidants in living organisms are not well understood either). They contain an iron atom in a heme, just as hemoglobin, the oxygen carrier in the blood.

To make potato juice for activity two, we will puree several potatoes in a blender. The puree works well, but the juice (with catalase) and the solids are still mixed. We can separate the juice with a small centrifuge. This part should be quite fun. You can also use cheesecloth.

For activity two, the adults should lead the children to build a protocol that resembles the following one.

In a test tube we will first add the enzyme source (potato juice or object to test for catalase activity). Next, we do something to the enzyme that could change its activity (change temperature, add a chemical…). Finally, we add hydrogen peroxide to see whether bubbling happens or not and when the reaction is done, measure "the extent of bubbling" (foam height for example). To make sure we can compare things together, each test must be accompanied by controls:

Example: Testing potato juice with vinegar

Tube "test": potato juice (10 drops) + vinegar (10 drops) + hydrogen peroxide solution (20 drops)

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Tube "control catalase": P.J. (10 drops) + water (10 drops) + hydrogen peroxide (20 drops).

Tube "control vinegar": water (10 drops) +vinegar (10 drops) + hydrogen peroxide (20 drops).

Measurement: Measure the height of the foam in each tube.

Prediction: Vinegar will speed up the reaction because vinegar makes bubbles with baking soda.

Result: ????

The controls let us compare the effects of vinegar and water on potato juice. It also lets us see whether vinegar has an effect of its own on hydrogen peroxide. The number of drops is purely indicative. What is important is to use the same quantities in the test and controls. As a hint to help you have varied results in a group, I can tell you that catalase is easily inhibited by cooking in hot water or in a flame, by the cold, by acid, by acetone

Equipment:

- Safety glasses for everyone
- Gloves
- Plastic pipettes
- Glass tubes (typically ½ in diameter, 3 in high)
- Glass tube holders
- Blender
- Centrifuge
- Centrifuge tubes
- Hydrogen peroxide solution 3%
- Potatoes, 4 or 5
- Eggs (3 or 4)
- Hot water
- Ice
- Salt, sugar, ice melt, baking soda, vegetable oil, bleach, acetone, vinegar…

as other possible sources: apple, piece of fresh meat, piece of cooked meat

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