Demonstrations in Soil Science

Contact:
Dr. John G. Graveel or Sherry Fulk-Bringman
Department of Agronomy, Purdue University
West Lafayette, IN 47907-1150
USA

Http://www.agry.purdue.edu/courses/agry255/brochure/brochure.html
DEMONSTRATIONS IN SOIL SCIENCE

Prepared by teachers in the Agronomy Department, Purdue University

Contents:

1. Measuring Soil and Water pH
2. Why is Rain Acid?
3. Testing Soils for Aluminum Toxicity
4. Soil Has a Charge
5. Chemical Movement in Soils
6. Nitrates or Nitrites in Water or Food
7. Exposing a Rainbow of Color: How Chromatography Works!
8. Soil Colors
9. Clay Properties
10. Soil Erosion
11. Earthworm Activity and Biology
12. Preserving Soil Monoliths and Specimens in Vinyl Plastic
13. Germination and Vigor of Seeds (Warm Tests/Cold Tests)
14. Quick Test to Determine Seed Viability
15. Phosphorus in Plants
16. Starch Goes to Sugar as Plants Use Their Stored Energy for Regrowth
17. Plant Growth Experiments

For more information contact:
Dr. John G. Graveel or Sherry Fulk-Bringman
Dept. of Agronomy
1150 Lilly Hall
West Lafayette, IN 47907
(765) 494-8060 jgraveel@purdue.edu

http://www.agry.purdue.edu/courses/agry255/brochure/brochure.html
1. MEASURING SOIL AND WATER pH

The symbol, pH, represents the negative logarithm of the hydrogen ion activity in a solution or suspension. It is used to express the acidity or basicity (alkalinity) of soils, water, and others solutions or suspensions. A pH of 7 is neutral; values above 7 are basic; and values below 7 are acidic. A pH of 6 is slightly acid and is common in many soils. A soil pH between 6 and 7 is best for growing most plants. When soil pH is below 5.5, the acidity may begin to cause serious problems for plants. Acid coal mine soils might be as acid as pH 3 and not permit plants to grow.

Each unit change in pH is a 10 fold change in acidity. Thus, pH 6 is 10 times more acid than pH 7 and pH 5 is 10 x 10, that is 100 times, more acid than pH 7.

Clean rain water is about pH 5.6. It is acid because the rain absorbs CO₂ from the air and produces carbonic acid (CO₂ + H₂O = H₂CO₃). Since the air has always contained CO₂, rain has always been acid. Modern day rain can easily be pH 4.6 or lower because it can be contaminated with sulfur oxides and nitrogen oxides which form sulfuric and nitric acids and give it additional acidity.

Materials

1. The pH of soil and water can be measured using indicator dyes. Indicator dyes can be purchased in a kit containing three dyes (Brom Thymol Blue, Chlor Phenol Red, Brom Cresol Green) which cover the pH range from 3.8 to 7.4. This range covers most natural systems including soils. These work on the principle that the indicator is one color when it is associated with H⁺ (as H-Indicator) in more acid systems and is another color when not associated with H⁺ (as -Indicator) in less acid, more basic systems. It is an intermediate color at intermediate pH values when partially H-Indicator form and partially in unassociated - Indicator form.

   e.g. for Brom Thymol Blue Indicator:
   
   \[ \text{H-Indicator} \leftrightarrow \text{H Indicator} + \text{-Indicator} \leftrightarrow \text{-Indicator} \]
   
   (yellow at pH 5.8)     (green at pH 6.6)   (blue at pH 7.4)

2. Collect a small amount of soil from your lawn or garden, and some water from a pond or from your source of drinking water.

Methods

Measuring pH in soil:

1. Put a small sample of soil in a spot plate hole (about 1/3 full). Add enough Brom Thymol Blue (BTB) to saturate the soil plus a few drops extra.

2. Tilt the spot plate back and forth to move the indicator solution through the soil, but tilt it gently so it doesn't get too muddy. Do this for at least a minute.

3. Roll the solution to one side and read its color from the top of the color chart; Blue = 7.4 or more, Blue-green = 7.0; Green = 6.6; Yellow-green = 6.2; Yellow = 5.8 or less.

4. If it goes yellow (i.e. for more acid soils) take another sample and try Brom Cresol Green (BCG). Read it from the bottom of the chart. It makes the same color changes but does so
between pH 3.8-5.4. Blue = 5.4; Blue-green = 5.0, Green = 4.6; Yellow-green = 4.2, Yellow = 3.8.

5. If you want to read pH between 5.4 and 5.8 (the range between these two indicators), then use Chlor-Phenol Red (CPR) with its color chart. The color change in Chlor-Phenol Red is more difficult to see since it is a yellow (pH 4.8) to red (pH 6.4) change. We suggest reading the color through a thin layer of the solution (rather than a thick drop) against the white background of the spot plate.

6. If you determine the pH of the soil to be less than 6.5, you can estimate the amount of limestone that would be required per acre to raise the pH to 6.5 from the table below:

<table>
<thead>
<tr>
<th>Texture</th>
<th>Light (low humus) soils</th>
<th>Dark (high humus) soils</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Soil pH</td>
<td>Soil pH</td>
</tr>
<tr>
<td></td>
<td>4.5 5.0 5.5 6.0 6.5</td>
<td>4.5 5.0 5.5 6.0 6.5</td>
</tr>
<tr>
<td>sandy loam to sand</td>
<td>4 3 2 0.5 0</td>
<td>10 8 5 2 0</td>
</tr>
<tr>
<td>silt loam or loam</td>
<td>6 5 3 1 0</td>
<td>8 6 4 2 0</td>
</tr>
<tr>
<td>clay loams or clays</td>
<td>8 7 5 2 0</td>
<td>10 8 6 3 0</td>
</tr>
</tbody>
</table>

Measuring pH of water:

1. You must be careful when measuring water pH because only a little contamination can change the pH. Use a small glass. Rinse it with distilled water several times; put in the water. Add enough indicator to be able to see the color. Don't get your fingers in it as your skin is very acid.

2. Read the pH of the water using the chart appropriate for the indicator dye added to the water. The specific way you read pH is the same for water samples as it is for soil.

Other samples on which you might measure pH:

- top soil from a flat farm field
- surface soil from an eroded hillside in a farm field
- surface soil (top inch) in a woods (nutrient recycling by decomposing leaves often gives the top inch a pH of 6 or higher).
- subsoil at various depths to see what the roots grow in
- water (rain, distilled, tap, stream, pond)

Precautions:

All three indicators (BTB, BCG, CPR) must be at the midcolor of their range when observed in the bottles. If any one is to the right of midcolor, add drops of diluted acid (very dilute HCl) and shake to adjust it. If any one is yellow, add drops of diluted base (very dilute NaOH) and shake until it reaches the midcolor.

Tap water and soaps are usually basic so rinse spot plates and glassware thoroughly with distilled water.
Your skin is very acid so don't get fingers into either the indicator solution or any water solutions.

Soils are very buffered and thus a little contamination isn't a problem with soils. Water is unbuffered so even a little contamination gives big errors in reading water pH.

Other techniques for measuring pH

1. **ColorpHast strips** are rigid plastic strips with a color pad on one end. The color does not bleed. The color changes are distinct and are compared to a color chart which is part of the small box the strips come in. Read the pH of any liquid or moist system with no contamination.

2. **A small, shirt-pocket pH meter** with digital read-out; reads liquids and moist systems very well; usually works quite well in soil suspensions. Should be checked against a standard pH 7 buffer solution and adjusted with a jeweler's screwdriver to read pH 7 if it is off. Powered by a small camera battery which is good for approximately 1000 hours. Sold by most general lab supply houses. One is listed below.

Sources of Materials:

- Indicator dyes - set of 3 with spot plate and instructions; covers pH 3.8-7.4. Available for $12 from the Purdue Agronomy Club, Department of Agronomy 1150 Lilly Hall, Purdue University, West Lafayette, IN 47907-1150.

- ColorpHast pH paper is available from Sargent-Welch phone (800) 321-8620, Catalog #S65271-2B for pH 2.5 to 4.5; -20C for pH 4-7; -200 for pH 6.5-10 at about $8.00 per box of 100.

2. WHY IS RAIN ACID?

Acid rain has been a major environmental concern. Its detrimental affect on streams and lakes and on fish populations within those waters is well documented. One would expect pure water to have pH of 7, neutral (Remember, pH’s less then 7 indicate acidity and pH’s more than 7 are basic or alkaline.) However, since air is approximately 0.03% carbon dioxide (CO₂), some CO₂ becomes absorbed into rain water and the pH drops due to the formation of weak carbonic acid.

Methods and Materials

1. Into a glass of drinking water add a few drops of Brom Thymol Blue solution (a pH indicator). It should give a light greenish-blue color indicating a pH of approximately 6.8. (Brom Thymol Blue is blue at pH 7.4, green at pH 6.6 and yellow at pH 5.8 or lower.) Many sources of drinking water have minerals in them that cause their pH to be near neutral, about 7.

2. Place a drinking straw or plastic tube into the water and blow. Your breath contains high amount of CO₂. What does the CO₂ of your breath do to the color? Why?

\[
\text{H}_2\text{O} + \text{CO}_2 \rightarrow \text{H}_2\text{CO}_3 \text{ (carbonic acid)} --- \text{Brom Thymol Blue turns yellow.}
\]

Atmospheric CO₂ in equilibrium with rain water gives rain a natural pH of about 5.6 (somewhat acid). But, today sulfur oxides and nitrogen oxides in the air from use of fossil fuels make rain even more acid, about pH 4.2 in some areas.

\[
\begin{align*}
\text{H}_2\text{O} + \text{SO}_x & \rightarrow \text{H}_2\text{SO}_4 \text{ (sulfuric acid)} \\
\text{H}_2\text{O} + \text{NO}_x & \rightarrow \text{HNO}_3 \text{ (nitric acid)}
\end{align*}
\]

There is little acid in rain. The H₂SO₄ and HNO₃ amount to only one-half pound of H⁺ per acre per year which requires about 25 pounds of limestone per acre to neutralize.

Acid rain may be a significant problem in other regions such as the northeastern part of the USA and eastern Canada where many soils are naturally acid, shallow and poorly buffered. A little additional acidity may be serious for them. Acid rain also has some benefits. It can provide most of the sulfur and a small part of the nitrogen that plants need to grow well.

The real acidity problem in most soils comes not from the acid rain but from the H⁺ produced by the microbial oxidation of nitrogen compounds, and from the natural leaching of basic cations from the soil over many, many years. These nitrogen compounds are produced by leguminous plants (alfalfa, clovers, soybeans) and of ammonium fertilizers (ammonium nitrate, anhydrous ammonia, urea) The acidity produced by legumes and ammonium fertilizers can require several hundred pounds of limestone per acre per year to neutralize.

\[
\text{NH}_3^0 + 2\text{O}_2 \xrightarrow{\text{microbial oxidation}} \text{HNO}_3 + \text{H}_2\text{O}
\]

(fertilizer ammonia) (oxygen) (nitric acid) (water)
3. TESTING SOILS FOR ALUMINUM TOXICITY

One of the most serious problems of acid soils is too much soluble aluminum. It is a problem in many humid and sub-humid temperate and tropical areas of the world. However, toxicity of aluminum to plants does not occur in all acid soils. This procedure describes a test for aluminum toxicity by bioassay. That is, we let plants tell us if the soil is toxic or not. Aluminum toxicity is indicated by a lack of root hairs and by the presence of short roots. (This same procedure can be used to test for the presence of other toxic substances, such as herbicides that inhibit root growth.)

Materials and Procedure

1. Find a good surface soil with a pH of at least 6. It will serve as a non-toxic control.

2. Collect some acid soils. Usually subsoils (below 12") are the most acid. The soils selected should be pH 5 or less. About half of the soils below pH 5 that we have tested exhibit aluminum toxicity.

3. On day 1, moisten the soils using a spray bottle and hand mix until each soil is moist enough for good plant growth but not muddy or puddled. Seal each soil in a plastic bag to allow moisture to equilibrate.

4. Also on day one, roll seeds of wheat, sorghum, or corn into wet paper towels and place them into a plastic bag and partially close opening in bag but do not seal it. Store in a warm place for about two days to germinate.

5. On day 3, place the soil in paper or plastic cups and plant five uniform, newly-germinated seeds in each (select seeds that have roots only a few millimeters long). Cover seeds with additional soil of the same type. Cover each cup with plastic held in place by a rubber band to keep the moisture from escaping. Set in a warm (about 25°C) place. Light is not necessary.

6. On day 5, dump plants out and look at the roots. Look for root hairs. Roots that hold soil have many root hairs. Absence of root hairs, the first sign of aluminum toxicity, results in clean roots with no soil adhering. More severe toxicity shortens the roots. Measure and compare root lengths between plants grown in the control soil and those grown in your acid soils. Calculate percent relative root length (%RRL):

\[
\text{average root length in acid soil} \\
\text{-----------------------------------------} \times 100 = \% \text{ RRL} \\
\text{average root length in control soil}
\]

7. Interpret your results. A % RRL value of 75% or less means the roots elongated only 3/4th as fast in the toxic soil and indicates the acid soil has rather severe aluminum toxicity. Values greater then 75%, but less than 90%, indicate less toxicity, but if root hairs are absent there may be reduced nutrient uptake. Values greater than 90% indicate little or no toxicity. Occasionally, the natural variability in seed may cause the roots to grow somewhat better in the acid soil than in the check soil.
One can add a gram of powdered CaCO$_3$ per 100 grams of soil to lime the soil, and repeat the experiment to see if liming eliminates the toxicity from the acid soil.

References:


4. SOIL HAS A CHARGE

How does soil keep fertilizers and pesticides from leaching out of the surface soil where they are needed? Soil behaves as an ion exchange column. This means that soil has a charge and can attract chemicals (ions) that have the opposite charge. Let’s determine if soil has a positive or a negative charge.

Materials and Methods:
A 6 volt battery, some copper wire, and a clay-water slurry will be used in this experiment. The clay can be purchased from the Purdue Agronomy Club, Department of Agronomy, 1150 Lilly Hall, Purdue University, West Lafayette, Indiana 47907.

1. Cut two lengths of copper wire about 8 inches long.

2. Attach one copper wire to the positive pole of the battery and attach the second copper wire to the negative pole. Be sure that the insulation is off the wire at the points of contact to the battery and in the clay slurry. (Any size battery will do, but a higher voltage battery works faster and better).

3. Place the ends of the wires in a flask or cup filled to the top with clay which has been mixed with water to the consistency of glue. If you decide not to use the clay supplied by the Purdue Agronomy Club make sure you use the stickiest clay you can find. An alternative to the clay could be a surface soil high in organic matter and clay.

4. After 10 minutes check to see whether the clay particles have moved to the wire attached to the positive or negative pole. Remember, unlike charges are attracted to each other. (Organic matter has the same charge as clay).

5. These ions are commonly added from limestone, fertilizers and acid rain to garden and farm soils:

   \[
   \begin{align*}
   &H^+ & \text{Hydrogen} \\
   &NH_4^+ & \text{Ammonium} \\
   &NO_3^- & \text{Nitrate} \\
   &K^+ & \text{Potassium} \\
   &Cl^- & \text{Chloride} \\
   &Ca^{++} & \text{Calcium} \\
   &Mg^{++} & \text{Magnesium} \\
   &SO_4^{=} & \text{Sulfate} \\
   &Mn^{++} & \text{Manganese}
   \end{align*}
   \]

Based on the results of the above demonstration, put a check behind those plant nutrient ions in the list above that will attach to the charge sites (exchange sites) on the soil. These ions will not leach out of the soil or will do so only very slowly.
Soils can do an excellent job of adsorbing many chemicals that are added to the surface or incorporated into the soil. However, not all chemicals are retained by the soil. Some chemicals move with the rain water that passes through the soil and thus, can contaminate ground water. As an example, when nitrate (NO$_3^-$) is added to the soil in fertilizers, or through wastes from septic systems or land disposal of manure it has the potential to move through the soil if not intercepted by plant roots or denitrified (returned to the air) by bacteria. If high concentrations reach the ground water, it can present a health hazard. Problems of nitrate movement into ground water are most serious in regions of sandy soils with shallow water tables in rainy seasons or when irrigation is used. Current evidence indicates that NO$_3^-$ is sometimes found in wells that are shallow and close to animal feeding lots, fertilizer dealerships, or home septic systems.

Why does nitrate (NO$_3^-$) move in soil and ammonium (NH$_4^+$), another form of nitrogen in soil, not move. Let’s conduct an experiment using a blue dye (methylene blue) which has a positive charge like ammonium and a reddish-orange dye (eosine red) that has a negative charge like nitrate.

**Materials and Methods:**
This demonstration uses colored organic dyes (one positively charged and the other negatively charged) to illustrate what happens when water soluble chemicals of different charge are placed in soil.

1. Use three glass tubes (columns) about 1 in (2.5 cm) in diameter and 10 in. (25 cm) long. (Size of tubes is not critical.)
2. Place a one-hole rubber stopper in the end of each tube with glass wool or cotton covering the hole on the inside.
3. Fill the tubes about 2/3 full with a loam or sandy loam surface soil and support the tubes in a rack.
4. Place a clear container below each column to collect solution passing through the soil.
5. In the first column of soil pour dilute methylene blue dye solution, it has a positive charge like ammonium, NH$_4^+$. (Dissolve approximately 1 gram of the organic dye in 1 liter of water to make the methylene blue solution.)
6. Into the second column pour a dilute eosine red solution, it has a negative charge like NO$_3^-$. (Dissolve approximately 1 gram of the organic dye in 1 liter of water to make the eosine red solution.)
7. Into the third soil column pour a mixed solution of the methylene blue and the eosine red.

**Questions:**
1. Does the blue or red solution come through the soil into the containers below?
2. Can you explain why the red dye moves through the soil and the blue dye is retained by the soil? (If you have not done the "Soil has a Charge" experiment in this brochure, you may want to conduct that experiment to help explain the results of this demonstration.)
6. NITRATES OR NITRITES IN WATER OR FOOD

Nitrogen is a key nutrient in plant growth. Nitrogen gas (N₂) makes up about 80% of the air we breathe and ammonium (NH₄⁺) and ammonia (NH₃) are commonly used in fertilizers. Nitrite (NO₂⁻) is used as a preservative in foods especially cured meats, nitrates (NO₃⁻) are common in soils, and organic nitrogen makes up enzymes, amino acids, and protein type materials in plants and animals.

Nitrate nitrogen is soluble and mobile in soils and may leach and contaminate groundwater or surface water. Normal water would be expected to contain some (NO₃⁻). In the United States the Environmental Protection Agency (EPA) has set 45 mg NO₃⁻/L (or 10 mg N/L in the nitrate form) as the maximum acceptable level for drinking water. Excess (NO₃⁻) in drinking water most often comes from leaching, excessive use of nitrogen, ineffective management of manure from concentrated livestock production facilities and septic systems.

While we normally eat NO₃⁻ in our food, new babies cannot tolerate high levels of NO₃⁻ during their first few days after birth. Methemoglobinemia, also called blue baby syndrome, occurs when certain bacteria in the stomachs of young animals and human infants convert ingested NO₃⁻ to NO₂⁻ (Brady and Weil, 1999). The NO₂⁻ interferes with the ability of the blood to carry oxygen to body cells. Unoxgenated blood is not red, hence the name blue baby syndrome.

Materials and Methods:

It is easy to test for NO₂⁻ (nitrites) and NO₃⁻ (nitrates) with a simple strip test. The plastic test strip has two pads on it. The zone at the very end indicates both "NO₂⁻ and NO₃⁻", while the other only reacts to "NO₂⁻." In the reaction NO₃⁻ is reduced to NO₂⁻ by a reducing agent. The NO₂⁻ reacts with N-[1-napthyl] ethylenediamine to form a red-violet azo dye.

The NO₃⁻ test is ideal for regular checks of drinking water, groundwater, surface waters, processed water, and wastewater. Test strips and the calibration chart can be obtained from Curtis Matheson Scientific Inc., 1225 N. Michael Dr. WoodDale, IL 60191. Order catalog no. 10020-1, EM Nitrate (NO₃⁻) Test, $24.60 per 100 strips.

Nitrates

A. In Water

1. Remove a test strip from the tube.
2. Dip the test strip into the solution to be tested for 1 second. Be sure both pads are wetted.
3. Remove the test strip, shake off the excess liquid, and after one-minute compare the color of the pad with the color scale on the container.

(If the pad at the very end turns red-violet and the other pad does not that means we have nitrates but no nitrites.)

B. In Soil

1. Thoroughly mix a sample of soil with the same quantity of distilled water and filter if necessary. (10 grams of soil with 10 milliliters of water).
2. Remove a test strip from the tube.
3. Dip the test strip into the solution to be tested for 1 second such that the pads are both wetted.
4. Remove the test strip, shake off any excess water, and after 1 minute compare the color of the pad with the color chart on the container.

Nitrites
Your spittle always contains NO$_2^-$ produced by your digestive system but no NO$_3^-$. 
1. Put some spittle on your finger and touch both pads to it.
2. Both pads test for NO$_2^-$ and should become equally colored.

References:
7. EXPOSING A RAINBOW OF COLORS: HOW CHROMATOGRAPHY WORKS!

The rate with which chemicals move through the soil is dependent on the chemistry of the compound applied, the strength of the interaction between the soil and the compound, and the flow of the water through the soil. The soil acts as a column that holds chemicals with varying degrees of strength. As water passes through the soil some compounds are held in the surface of the soil and others move rapidly through.

We can illustrate how compounds can be separated into individual components by observing what happens when water interacts with a black line placed on a piece of filter paper.

Methods and Materials

1. Take a filter paper disk (approximately 3 inches in diameter) and place a straight black line on the paper approximately one-third of the distance from the edge. Be sure to use a marker with water-soluble ink. See diagram below:

   ![Diagram](image)

   A B C

2. Take the edge of the filter paper at "A" and roll the paper back. Do the same with the filter paper edge at "B." Staple or paper clip these two edges together. You now have a modified tube.

3. Place the edge of the paper at "C" into a beaker of water. Be certain that the water level is below the ink line. (Deionized or distilled water is preferred, but not required.)

4. Watch the water rise on the filter paper by capillary action. As it moves past the line, note how the colors that compose black separate into the colors of the rainbow.

Explanation

The black line on the filter paper is composed of many colors. However, each color has a different degree of solubility in water and in its attraction for the filter paper. Thus, as the water rises, each color can be seen separately. The process of separating chemicals by using a support medium (the filter paper) and a separating solution (water) is called chromatography. Police investigators use chromatography to reveal new evidence and scientists use the technique to discover new compounds for medicines or for isolating genetic material. In nature, soil behaves as a chromatography column. As compounds are added to the surface of the soil, they move at different rates through the soil depending on their solubility in rainwater and their attraction for the soil particles.
8. SOIL COLORS

Color is an indicator of many soil characteristics. Organic matter from the decay of plants and animals causes soils to be dark in color, even black, if enough is present. Iron compounds provide most of the red, brown, and yellow colors in soils. The presence of these colors indicates good aeration or good soil drainage. Poorly drained (poorly aerated) soils have dull colors, grey and blacks, whereas those that have good drainage have brighter colors of iron in its oxidized state, $\text{Fe}^{3+}$. This characteristic allows soil scientists to predict which soils are normally wet even when observed in a dry period. This is helpful in selecting locations for homes, other buildings, or waste disposal sites.

You can demonstrate this change in color when soil organisms can't get $\text{O}_2$ from the air and begin to reduce iron, causing it to lose its bright color. Follow these procedures to cause a red or brown soil to turn grey.

Materials and Methods

1. Place about an inch of red, brown or tan subsoil into each of two bottles or flasks. Add enough water to cover the soil about one inch deep.

2. Seal the first flask by snapping a balloon over the bottle top. This soil has been made anaerobic (oxygen supply eliminated) but it has no energy source for the anaerobic organisms.

3. Add 1/4 teaspoon (1.0 ml) sugar to the second flask (you may wish to do another bottle substituting a teaspoon of finely ground alfalfa meal) as energy and carbon supply for the microorganisms. Also add a small pinch of a poorly drained soil (this serves to inoculate the system with plenty of anaerobic organisms). Seal it with a balloon snapped over the bottle top and set both bottles in a warm place.

4. Place a teaspoon of dark topsoil in a ceramic crucible and heat over an open flame until it stops smoking. It is okay to heat until the crucible glows red. Do this on several soils of different color.

Observations and Expected Results

1. Bottle one will probably not change or change very slowly because the subsoil material usually does not contain enough organic matter (energy source) for the microbes.

2. The anaerobic microbes will begin to work in bottle 2. After a few days you can see the bubbles of CO$_2$ rising in the water; also the balloon may begin to fill with gas.

3. After 10 days the soil in bottle 2 should have turned grey (reduced) or at least begun to do so. This is what happens in water logged soils especially when there is enough food for the organisms. It is why the subsoil of "poorly drained" soils is grey and why "somewhat poorly drained" soils have grey mottles, or splotches of grey.
4. When you open the flask with the grey soil it will probably have a bad odor from the organic acids formed by anaerobic decomposition. Pour the water off into a bottle and let it stand with a lid on loosely to allow oxygen to enter. This water will contain some of the iron reduced by the organisms because reduced iron is more soluble in water than the oxidized iron. After a few days the Fe\textsuperscript{2+} in the water as FeO should form Fe\textsuperscript{3+} as Fe\textsubscript{2}O\textsubscript{3} which is red and quite insoluble. Red and brown iron concretions in soil form when reduced (Fe\textsuperscript{2+}) iron in solution moves to a zone with more oxygen and accumulates in the oxidized, (Fe\textsuperscript{3+}) form as Fe\textsubscript{2}O\textsubscript{3}.

5. The soils you heated to a high temperature in the air will have the organic matter burned away. The black color in surface soils is mostly from organic matter (humus). Burning the humus from soils by heating them red hot over a flame or on a stove will reveal the iron color (tan, yellow, brown, or red) that was hidden by the humus.
9. CLAY PROPERTIES

Clays are very important in agriculture and in a wide variety of other applications including such things as manufacture of ceramics and sealing ponds or waste sites. This exercise demonstrates one of the important properties of clays, their ability to absorb water and expand to many times their volume. This characteristic of clays is valuable when used to seal a basement wall or the bottom of a water reservoir, but it causes problems when clayey soils expand and push against walls or other structures.

Materials

Flat open dishes such as petri dishes.
Paper cups, 4-6 oz.
Graduated cylinder for measuring water volumes.
Mixer such as a food blender or a milkshake blender.
Clays: montmorillonite and kaolinite

Methods

Three techniques can be used to show differences in clay expansion as it is wetted.

1. Place a tablespoon (about 30 ml) of dry montmorillonite into one paper cup and a similar amount of kaolinite into another. Add water to each in about 25 ml increments and stir vigorously. Record the amount of water used and continue to add water and stir until the suspension of clay and water flows easily like thick soup or oil. You should note a big difference in the amount of water the clays absorb before they flow as liquids.

2. Start the blender stirring with 500 ml of water in it. Add two tablespoons (60 cc) of kaolinite to the water. Observe that the resulting suspension of clay and water is liquid. After cleaning the blender begin again with 500 ml of water and this time add two tablespoons of montmorillonite clay, but add it slowly, sprinkling in a few cc at a time. Continue this until the blender begins to slow or choke down or until all the clay is used. You should note that one of the clays absorbs huge amounts of water and still behaves as a solid or thick liquid. Which clay?

3. Prepare a thick paste or slurry of each clay by mixing with water until it behaves as a viscous liquid, just barely flows. Fill two flat, shallow dishes with each of the clay-water mixes. Cover one of each with a lid or plastic wrap and allow the other one to dry in the open air. Note the differences in cracking and the amount of clay left after the water evaporates. Compare the remaining clay with the full dish that has been protected from water loss. Which clay had the greatest reduction in volume as the water evaporated?

1) Montmorillonite and kaolinite clays can be ordered from the Purdue Agronomy Club, Department of Agronomy, 1150 Lilly Hall, Purdue University, West Lafayette, IN 47907-1150
10. **SOIL EROSION**

Erosion, the loosening and movement of soil by wind and water, presents a significant limitation to sustained agricultural production world-wide. Past civilizations have collapsed as their soils, once deep and productive, washed away leaving shallow infertile soil behind. World-wide, during the past 50 years, we have degraded 5 billion acres of land, most of that by wind and water erosion. In the United States about 4.5 billion tons of soil are moved annually by soil erosion.

The effects of erosion are twofold: first, erosion reduces soil productivity by removing soil nutrients, degrading soil structure, and decreasing soil water holding capacity and plant rooting depth. Second, sediment the end product of erosion, is a major source of water pollution. Among the environmental problems caused by sedimentation are the silting-in of stream channels and reservoirs, restricted stream flow that increases flood hazards, and impaired water quality due to sediment transport of nutrients and pesticides. In this experiment you will design a demonstration to illustrate the effect of soil properties on soil erosiveness.

**Materials and Methods:**

One of the primary goals of this project is to construct a soil erosion demonstration that provides students with insight into practices that protect the soil and minimize the impact of erosion on water quality.

The procedure used in this demonstration is adopted from Beck (1984). The materials used are easily purchased from any lumber yard or hardware store. The materials list can be found in the section on Construction Materials.

1. To assemble the soil erosion display model, lap joints were cut at each end of the four pieces of lumber stock with dimensions 2 x 4 x 36 in. These four pieces formed the base. Next, two holes were drilled vertically at each corner through the lap joint so that the 3/8 x 2 1/2 in. carriage bolts could be inserted and secured with the wing nuts to form a sturdy base.

2. The four uprights were positioned inside the corners of the assembled base and two horizontal holes were drilled through the base and through each upright. The holes were drilled from the side of the base so that they passed through the 4 in width of the base and then through the 2 in thickness of the upright. Two carriage bolts (3/8 x 6 in with wing nuts were used to secure each of the uprights.

3. The two supports for the plastic pipe halves required the cutting of half circles along the upper side of each piece. The cuts were 4 in diameter and spaced evenly across each piece. The top of the front support was secured 14 inches above the bottom of the base by two carriage bolts inserted through each of two horizontally drilled holes in each upright that matched the two holes drilled in each side of the front support. The back support was secured to the back uprights in the same manner, however the top of the back support was positioned 16 inches above the bottom of the base.

4. The two supports for the plexiglass tray were secured to the uprights 4 inches below the top of each upright. Two horizontal holes were drilled through the 2 in thickness of each of the uprights and the 2 in thickness of each end of the tray supports. The last eight carriage bolts (3/8 x 4 in) and wing nuts were used to fasten these tray supports in place. A diagram of the assembled wooden portion of the display is shown in Figure 1.
5. The plexiglass tray was glued together to form a tray 13 x 24 x 4 in. which rested upon the wooden supports placed 4 from the top of the uprights. Four rows of holes were drilled through the bottom of the plexiglass tray. These four rows were positioned directly above each of the four plastic pipe halves resting below the plexiglass tray. The holes were slightly smaller than the rubber serum stoppers so that a water tight seal was obtained when the serum stoppers were inserted into the holes. The holes were alternate so that two rows of holes were drilled with 12 holes per row. After the serum stoppers are in place, hypodermic needles should be pushed through the stoppers from the inside of the tray. When water is added to the tray "raindrops" will form when passing through the needles.

6. The trays contain soils of varying textures or management practices. For example: Tray 1 - sandy texture, Tray 2 - silty texture, Tray 3 - clayey texture, and Tray 4 - a silty textured soil with a thin layer of mulch applied. There are a wide variety of management practices to try such as growing soybeans in the trays under conventional tillage and no till. The possibilities are endless.

7. Next "rain" is applied to the soils by adding water to the plexiglass tray.

8. Sediment rolling off the trays is collected in buckets at the end of each tray and used to measure soil loss. Observe the amount of sediment removed from each tray and discuss the impacts of these losses on soil management and water quality issues.

For people interested in other techniques for measuring soil erosion I recommend the "Bottle Hydrology: Runoff/Erosion Model" developed by Kevin Fermanich at the University of Wisconsin. Figure 2 is a runoff/erosion model constructed from 4 two-liter plastic soda bottles and at least one bottle cap. For more information about this demonstration contact: Department of Soil Science, University of Wisconsin-Madison, 1525 Observatory Drive, Madison, WI 53706.

**Construction Materials**

- 4 - 2 x 4 x 36 in: Base
- 4 - 2 x 4 x 30 in: Uprights
- 2 - 2 x 4 x 28 in: Supports for plastic pipe halves
- 2 - 2 x 2 x 28 in: Supports for plexiglass tray
- 8 - 3/8 x 2 1/2 in carriage bolts
- 16 - 3/8 x 6 in carriage bolts
- 8 - 3/8 x 4 in carriage bolts
- 32 - 3/8 wing nuts
- 1 - 24 x 30 in sheet of plexiglass cut to give 5 pieces:
  - 1 - 24 x 13 in
  - 2 - 24 x 4 in
  - 2 - 13 x 4 in
- 2 - 4in x 6ft plastic pipe cut in half to make 4 trays
- 48 - hypodermic needles 16 gauge
- 48 - rubber serum stoppers

**References:**

Earthworms are the most important macroanimal present in the soil. They burrow through the soil eating organic matter and ingesting soil. Earthworms eject large quantities of soil and partially decomposed organic matter that enhances the fertility and productivity of the soil. The holes left behind by earthworms serve to increase the aeration and drainage of the soil. Earthworms multiply rapidly in cool, moist (but not too wet) soils if there is an adequate food supply such as grass clipping, vegetable peelings, and compost.

Part 1: A study of the ability of worms to penetrate and mix soil.

Materials and Methods:
1. Find four healthy worms (smaller earthworms are preferable over the large nightcrawlers).
2. Obtain a dark colored (high in organic matter) topsoil and a light tan or brown colored subsoil.
3. Dry and powder the two soils.
4. Fill the bottom 1/2 of a jar with the light colored subsoil and the next 1/4 with dark colored topsoil.
5. Mix a small amount of food for the worms into the topsoil - grass clippings, leaves, etc.
6. Moisten the soil by adding small amounts of water to the top until the wetting front almost reaches the bottom. If your soil was uniformly fine it will wet very slowly. Be careful not to get it overly wet or saturated.
7. Drop the worms in and put the lid on loosely (to permit air exchange).
8. Worms prefer darkness so make a sleeve of opaque paper or aluminum foil to drop around the jar.
9. Check the activity of the worms daily or weekly to see how much soil mixing they do and how many channels they make.
10. Later, remove the lid and let the soil dry down for a week or two. Then add water and note how rapidly it enters the soil and wets it to the bottom, in contrast to when you first wetted the soil (earthworms improve infiltration of rain which then reduces runoff and erosion).
11. Carefully break the jar to remove the soil as undisturbed as possible (do it with gloves and goggles on so you don't get hurt by the glass). Slice the soil horizontally or carefully break it apart to study the size and number of worm channels.

Part 2: Start a worm farm to study worm biology.

Materials:
1. Find a location with earthworms in the soil - a moist, shady spot, by leaf or compost piles.
2. Find or make a box at least 6 in. (15 cm) deep x 12 in. (30 cm) x 20 in. (50 cm).
3. Fill the box almost full of soil from the location where your worms are found. Dig for extra worms to put in the box, a few dozen would be a good start.
4. Mix in several handfuls of rotten leaves; compost or similar food and be sure the soil is
moist (not wet).

5. Cover the soil with a moist cloth and cover the box loosely with a plywood board to
reduce evaporation. Put in a shady place.

6. Each week lift the cloth and sprinkle a little cornmeal or compost on the soil surface, and
remoisten the cloth and the soil surface. You'll soon learn when the soil is too wet or too
dry.

Activities:

1. Harvest worm eggs:
   In 3 or 4 weeks there should be some worm eggs in your box. Egg capsules are lemon
colored and about 3 millimeter in diameter. Each 3 or 4 weeks you can dump out the soil
onto a newspaper sheet and sift through it to collect the eggs. Collect them carefully on
some moist paper in a small container.

2. Watch worms develop:
   You could start a new small breeding box with the eggs. It could be dumped out every few
days to find whether the eggs have hatched, how fast the worms grow, etc. Some worm
species mature rapidly (about 90 days) while others take nearly a year to mature. The eggs
may take 3 weeks to 3 months to hatch, depending on species and conditions. Also worms
are hermaphrodites (each worm is both male and female) so all worms produce egg
capsules.

   *Lumbricus terrestris*, the common "nightcrawler" is a large worm and goes deep into the
soil. *Apporectodea tuberculata* and *Apporectodea trapezoides* are smaller, pale pink
earthworms that stay near the surface. Which species do you have?

3. The nightcrawler likes deeper soil and surface-feeds taking plant tissue down into its
burrow. Fill a large bucket with moist soil and add a few night crawlers. Put 3 or 4 small,
separate piles of different residues on the soil surface. Residue should be cut to the same
particle size. You might use soybean, corn, oak leaf, or other organic sources. Which
residues do the worms prefer? By experimenting with additional foods one may be able to
deduce what makes food attractive to a worm.
12. PRESERVING SOIL MONOLITHS AND SPECIMENS IN VINYL PLASTIC

Soil profile samples (monoliths) are four-foot vertical soil samples taken and preserved in their natural state. They are easy to take especially when vertical soil cuts are available from road cuts, basement excavation, and soil judging contests. Monoliths are excellent for use in night classes, winter schools and all educational occasions when you can't get out to see the soil in situ. A video, V-AY-12 is available from Purdue University on how to take soil monoliths. Contact the Media Distribution Center at (765) 494-6794 for purchase of this video.

Materials and Methods:

1. Make trays 1" deep x 4" wide x 46" long of 16 gauge galvanized sheet metal. Welding must seal corners or tight riveting so liquid plastic fixative will not leak out.

2. Order acetone* from a chemical supply house, drug store, or see your high school chemistry teacher. Buy the cheapest grade they have. Danger!!!! Acetone is volatile and inflammable! Handle like gasoline and always work in a well-ventilated area.

3. Order powdered vinylite plastic from the Purdue Agronomy Club, Department of Agronomy, 1150 Lilly Hall, Purdue University, West Lafayette, IN 47907-1150. Recent price, $3.00/lb. One lb (0.5 Kg) will do three to four monoliths.

4. Select soils that teach a lesson or that are good representatives of the various soil types.

5. Force the tray into the face of the vertical soil cut by tapping the backside of the tray gently with a sledge hammer or heavy axe. (Place a board against the back of the tray to prevent denting.) Keep constant pressure on the tray with your hand (at the top) and the toe of your boot at the bottom. The tray must never spring back or loosen after you start taking the monolith.

6. Dig around the tray with a large knife. Break the top part of the tray full of soil loose from the cut by slicing down in front of the tray with a sharp shooter spade, then break the sub-soil loose with your hands. It is best to have two persons to hold the soil in the tray while it is being broken loose. Steps 5 and 6 take less than 30 minutes when a moist soil cut is available with few stones or hardened layers.

7. With your sample now taken, lay it horizontal and pick off the excess soil with a knifepoint. Don't leave knife marks. Pick the soil off so the natural structure is exposed. Blow off all loose particles. Mark the horizon boundaries on the tray edge.

8. Let the profile sample air dry.

9. Dissolve vinylite in the acetone to make a thin solution (1/2 pound (1/4 Kg) of vinylite in about 2 qts (2 liters) of acetone. Vinylite goes into solution very slowly.

10. Drip the thin solution of vinylite onto the dry profile sample. Soak the profile well. Use at least a quart. If too much is added the surface appears glassy.

11. Let dry, the acetone evaporates and the plastic becomes solid, then the monoliths are ready for use. The plastic has become the glue that permanently holds the monolith in the tray.

12. The same vinylite solution works well for preserving other soil samples that illustrate worm holes, soil structure, and other features you wish to preserve.
13. After some years of use, a monolith becomes dusty and dull in appearance. To “refresh” the appearance, re-treat the surface with a dilute solution of the plastic in acetone.

*Acetone is the ketone of low molecular weight and it evaporates rapidly. A more complex ketone, 2-butanone, also works and is better because it evaporates slower and is less apt to leave a glossy finish on the soil, however it is more expensive than acetone.
13. GERMINATION AND VIGOR OF SEEDS (Warm Tests/Cold Tests)
from M. Rathert and J. J. Vorst

Viable seeds will germinate if placed under the proper conditions of moisture, temperature and oxygen. After germination, seeds differ in their rate of growth, depending on the physical condition of the seed, and the environmental conditions present. Damaged or weak seeds planted in cold, wet, disease infested soils seldom produce healthy plants, even though the seed may have had a favorable germination percentage. When seed is purchased, the germination percentage expressed on the seed tag is the result of a warm germination test conducted under nearly ideal conditions. A cold vigor test more nearly approximates the conditions under which many seeds actually germinate in a field. The same germination test is also used to determine the germination percentage of lawn grass and vegetable seeds.

Low quality seed may be caused by poor harvesting and storage conditions, rough handling during cleaning, processing and shipment, and disease or insect infestations.

The warm germination test determines the ability of seed to produce a stand under favorable environmental conditions. When compared to actual field performance, the warm germination test overestimates field emergence.

The cold vigor test attempts to more closely simulate field conditions at planting time. As planting date becomes progressively earlier, germination and emergence conditions become harsher because soils are cooler and more damp. Cool, damp soil conditions are ideal for development of seed and seedling diseases, especially on slowly growing seedlings.

To determine seed viability and vigor, the following tests may be conducted. Soybean seed is used because the seeds are large and differences in viability and vigor are easy to determine. Seed can be damaged by dropping them from a 3 meter height onto a hard floor. Dropping the seed 3 or 4 times results in damage levels similar to those of soybeans that are improperly handled during processing.

Materials and Methods
A. Warm germination test

1. Wet a sheet of germination paper (or non-chemically treated absorbent paper) with cold water, and place on a sheet of white butcher paper.

2. Count out fifty seeds from each seed lot and place them evenly in five rows of ten seeds on the germination paper.

3. Cover the seed with a second sheet of wet germination paper and fold the bottom of all three sheets of paper over about 3 cm.

4. Roll the butcher paper with the germination paper inside loosely, but tight enough to hold the seeds in position on the paper. Place a rubber band around the middle, and label the roll.

5. Repeat the above procedure for each seed lot.
6. Place both rolls in a 25°C (77°F, i.e. about room temperature) moist environment for 7 days.

B. For cold vigor test

1. Place 2.5 cm (1 inch) of vermiculite in each of two boxes approximately 20 x 30 cm in size. (Plastic shoe boxes available at any discount store work well.)

2. Place 50 soybean seeds from each seed lot on top of vermiculite in five rows of ten seeds.

3. Place 2.5 cm of vermiculite on top of seeds.

4. Water well with tap water, and place a lid on the box, and label the box.

5. Repeat the above procedure with each seed lot.

6. Place the boxes in a 10°C (50°F, about refrigerator temperature) environment for 7 days.

7. After 7 days, place in a 20-25°C (room temperature) environment for 5 days.

C. The data

1. For the warm germination test, count the seedlings that have germinated and have no abnormal or deformed parts. Multiply this number of seedlings times 2 (because you only had 50 seeds) to obtain the warm germination percentage.

2. For the cold vigor test, count the number of seedlings that have emerged. Multiply this number of seedlings times 2 to obtain the percentage of plants that would be expected to establish under field conditions.
14. QUICK TEST TO DETERMINE SEED VIABILITY

We need to know whether seed we plan to use is alive. Farmers often use last year's grain (beans, wheat, oats, etc.) for seeding the new crop. The tetrazolium test is used to furnish a quick estimate of seed viability. It also illustrates to students which part of a seed is "alive" and which part is simply storage tissue. Because the seed is not subjected to all of the environmental factors that reduce germination where planted in the field, the actual number of seeds that germinate and emerge in the field is less than predicted by this test.

Tetrazolium solution is colorless but respiring (living) tissue gives off hydrogen which turns the tetrazolium bright pinkish red. A bright pink color indicates healthy tissue, whereas a dark red color indicates embryos that have been bruised, frozen or damaged in other ways. Buy tetrazolium (2,3,5 triphenyl tetrazolium) from a chemical supply house (e.g. Fisher item #T 413-10) and make it up in water within the range of a 0.1 to 1.0% solution. The 0.1% works well for corn; higher concentrations are better for soybeans and small seeds. Add a drop of detergent to the solution to wet the seeds better.

**Methods and Materials**

The tetrazolium test is conducted on corn and soybeans by using the following procedure:

1. Moisten absorbent paper in a petri dish with tetrazolium solution.
2. Split 10 presoaked corn kernels with a sharp blade and lay the flat cut side onto the wet tetrazolium paper. The bean seeds can be laid onto the wet paper without splitting. The corn kernels should be presoaked 6-12 hours, and the soybeans 2-4 hours.
3. Cover the dish and let react for two hours.
4. Evaluate the seeds. The embryo of the corn and the soybeans should turn bright pink if the seeds are viable (alive). The degree and intensity of color can be related to the vigor of the seed. Healthy tissue is indicated by a bright pink color, damaged tissue by a dark red to black color, and dead tissue by a white color. Seeds with embryos which do not stain in two to three hours are dead seeds and would not germinate under any conditions. Seeds which exhibit a bright pink color throughout the entire embryo will normally germinate and produce healthy plants if the seed is exposed to ideal germination conditions.

Refer to the attached sheet to determine the vigor of the seed. The dark areas indicated by arrows refer to damaged tissue and will be stained dark red to black. Seed in class 1 will be bright pink. Sometimes, if the stain has not had time to reach the center of the seed, the center will still be white. If the outer edges are bright pink, the seed is viable. Seed in classes 2 through 5 are damaged, but the damage is not severe enough to cause the seed to be nonviable. While these seeds will germinate, they may not produce healthy plants under cool, damp field conditions.

Seed in classes 6 through 8 are severely damaged and are considered nonviable. These seeds should be counted as dead, and added to those that absorbed no stain to determine the number of nonviable seed.

To determine the germination percentage, multiply the number of seeds in classes 1 through 5 by 10. You may wish to conduct multiple tests of each seed lot to increase your accuracy.
References


15. PHOSPHORUS IN PLANTS

Phosphorus (P) is one of the essential elements in plants and animals. It is important in energy transfers in plants. Phosphorus fertilizer additions are often needed to obtain vigorous plant growth. Diammonium phosphate, $(\text{NH}_4)_2\text{HPO}_4$, is a common fertilizer that supplies phosphorus and nitrogen. Tests of plant and soil phosphorus content are necessary in well-managed plant production systems.

Plants can be tested for P using a color test. Phosphate in plants, $\text{PO}_4^{3-}$, can be extracted from leaves with a dilute acid and combined with molybdate to form a blue solution. The phosphate ions are extracted from the fresh plant and reacted with 12 molybdate ions, $\text{MoO}_4^{2-}$, which form a cage around each phosphate producing a phospho-molybdate complex.

The phospho-molybdate complex ion is almost colorless. However, when chemically reduced, the complex ion turns blue. More phosphate ions in the plant sap produce more complex phospho-molybdate ions and the solution turns darker blue.

**Materials and Methods**

1. **Molybdate reagent:** used to extract the P and form the complex.
   
   Dissolve 1 g of ammonium molybdate in 250 ml of distilled or deionized water. Slowly and carefully add about 16 ml of concentrated hydrochloric acid (HCl) with constant stirring. (This reagent is only useful for a few months. See the note below.)

2. **Reducing reagent**
   
   Tin compounds are often used to chemically reduce the complex.
   
   a. Powdered stannous (tin) oxalate (or) a Piece of "solid-core" solder (solder contains tin), works fine.

**Procedure**

1. Cut fresh plant tissue into very fine pieces, with a razor blade.

2. Place about 1/2 teaspoonful (2 ml) of the tissue into a large test tube or small bottle.

3. Add 20 ml of molybdate reagent.

4. Shake vigorously for 1 minute.

5. Add one fleck of stannous oxalate, about the size of a pin head and stir. Don't add more as it may result in a discolored greenish solution. (An alternative is to stir the solution with a piece of solid-core solder, which will provide enough tin to cause the reduction of the complex.)

6. Compare the color to the following qualitative descriptions:
   
   Colorless or Yellow          very deficient in P
   Green or Light Bluish Green   deficient in P
   Light Blue                   medium in P
   Medium Blue                  adequate in P
   Dark Blue                    abundant or excess P
If a house plant is deficient in P you can add a small amount of fertilizer containing P such as 18-46-0 (about 10 to 15 pellets) or a house plant fertilizer. Then water the plant and in a few days check again to see if the P content of the plant has improved.

A dark blue color means that your plants are getting all the P they need and probably an excess. You may be over fertilizing. Over fertilizing fields or garden soils is wasting money on excess P fertilizers and could be polluting the surface water by run-off water or by erosion.

Note: The molybdate solution is usually good for several months before a fresh batch must be made. To determine whether it is still good add a speck of stannous oxalate to 10 ml of the solution (or stir it with the solder). If it turns blue (without P present) it is no longer useable.

Studies You Can Do

1. Test plants growing in places that might have abundant P (well fertilized house or garden plants, plants growing near animal feed lots and barns, dark green healthy clumps of grass on lawns, etc.) and compare with plants growing on sites that might be P deficient (eroded spots, light colored soils, unfertilized gardens, etc.)

2. Grow plants in soil with and without addition of P fertilizer and test their tissue for adequacy of P.
16. STARCH GOES TO SUGAR AS PLANTS USE THEIR STORED ENERGY FOR REGROWTH

Suzanne Cunningham and Jeffrey Volene

Starch is the major storage form of energy in many plants. We often harvest the starchy part of plants for food or feed (corn grain, potatoes, etc.). Starch is a simple polymer chain consisting of hundreds to thousands of single glucose sugar molecules linked together. Plants not only make starch to store energy but they also can produce enzymes which convert starch back to sugar when they need it. For example, the starch in a corn seed goes to sugar as the seed germinates so the new seedling will have energy to grow until it can produce its own sugars by photosynthesis. The starch stored in an alfalfa root is converted to sugars after the alfalfa is cut so that the clipped plant will have enough energy to regrow.

Materials and Methods:

There is a simple iodine test for starch. The long starch polymers are coiled like a spiral staircase and there is space inside the coil just perfect to trap iodine molecules. Thus, iodine solution added to starch produces an intense blue-black color from the trapped iodine molecules. However, as enzymes decompose the starch to simple glucose molecules there is no longer a space in which to trap iodine molecules.

1. The test solution is made from potassium iodide (KI) and iodine (I₂). Exact composition is not important but about 2 grams of KI and 0.2 grams of pure I₂ in 100 ml of distilled water (or if your iodine is already a solution its final concentration should be about 0.2%).

Alfalfa

2. Dig alfalfa roots and slice them off like cutting the top off of a carrot. Add the iodine test solution to the cut area, wait a few minutes, wash off with water and observe the iodine-stained area.
   a. When alfalfa is in bloom, it has stored lots of starch and is ready to be cut.
   b. One week after cutting much of the starch has been converted to sugar for regrowth energy.

Corn

3. Start corn seeds germinating and then slice open individual seeds every few days to observe starch depletion as enzymes convert it to sugar for the young seedling.
17. PLANT GROWTH EXPERIMENTS

Plant growth experiments or demonstrations can be performed to illustrate plant response to many environmental variables or test experimental hypotheses. There are many ways to establish such experiments. The following suggestions were developed to guide student projects.

1. In pot experiments use large pots and lots of soil. This gives a more natural rooting volume, makes weighing and mixing the soil and fertilizer, lime, etc. easier and more accurate and you don't have to water as often.

2. Keep the population reasonable. Four corn plants in a gallon pot is many times more dense than normal field population.

3. Establish a wide range or contrast in the experiment. With nutrient response experiments it is good to start with soils known to be very low in the element of interest. For example soil from a well drained forest or other uncultivated area is often very low in N, P and K, especially if one uses the soil just below the top few inches of "A" horizon.

4. Have or add adequate levels of all nutrients except the one you are studying.

5. For initial experiences of students stick to treatments that can be expected to show noticeable differences, e.g. the N response of corn or tomatoes on light colored soils. Most experiments designed to show effects of micronutrients will probably fail because the seed and even a poor soil often provide enough for short term experiments.

6. Replicate the experiment at least 3 times to allow meaningful analysis of the results and to demonstrate variability in the best of experiments.

7. Water by weight. Have the soil hand-mixed to the correct moisture when you put it in the pots to start the experiment. Weigh each pot with its soil. Each time you water bring the pot back to its original weight. This prevents the common problem of over watering.

8. Greenhouse (in door) experiments should be planned to last a few weeks, 4-8. Plants are growing under abnormal conditions and become stressed as they become large due to limited root volume and crowded conditions.

9. Measure several variables e.g. date of emergence, height at weekly intervals, number of leaves showing, deficiency symptoms, fresh and dry weight at harvest.

10. Provide daylight if possible. It is difficult to provide sufficient light artificial light for normal growth. Remember daylength is important to growth of many species.