Soy in Food: What is that doing in there?
Macromolecular Composition of Field Crops
Teacher Instructions

Description:
This laboratory exercise will allow students to explore the various macromolecular compositions of field crops (corn, soybean, wheat). All of these plants contain the macromolecules protein, lipids, and carbohydrates. These molecules are large and complex, often composed of many subunits (hence, macro). They serve various functions in sustaining plant life itself, in providing nutrition to animals when they are included in animal feed, and nutritional as well as product-specific functions in the foods we consume daily. Therefore, this activity can be integrated into various classroom activities and it is suggested that teachers seek to contextualize their introductory discussion within their classroom's area of expertise.

Materials:
Soybeans, wheat, and corn
Soy flour, vital wheat gluten flour, corn meal
Iodine solution and/or enzymatic kit
Biorad protein kit and/or send off of protein analysis
Mortar and pestle
Ethanol or acetone
Cheesecloth
Bowls
Spoons

Day 1:
***Discussion* –** general guidelines, but can be modified to fit classroom

- Describe macromolecules and their function within cells
- Ask students to speculate about the relative amounts of macromolecules in corn, soy, and wheat
- Ask students if they have heard of certain proteins/allergens associated with these crops
- Ask students if they are familiar with different types of carbohydrates, lipids, and proteins.
- Ask students if they know examples of foods associated with these macromolecules.

***Experiment* – Gluten extraction**

- Students will attempt to extract gluten from the three flours (corn meal, soybean flour, vital wheat gluten or whole what flour)
- Break students up into teams of three. Each student in the group is responsible for one of the flour types.

- Each group will need three small bowls, three spoons, one cup of each flour, water, and three 6x6 inch swaths of cheesecloth. The specific amounts listed here are not essential, getting close is sufficient.
- Have each student place one cup of flour into each of three bowls and add 1 cup water
- Have students stir vigorously with a spoon, they will likely have to use their hands to knead as the gluten becomes rubbery. The other mixtures will remain soupy.
- Students will transfer the mixtures to the cheesecloth, and wrap to strain. Eventually the corn meal and soy flour will be rinsed away with water, leaving nothing left.
- Rinse cheesecloth bags under running water. If there is not access to running water, simply replace the water in the bowls that were previously used and continue washing. The amount of gluten remaining in the cheesecloth will by much smaller than the volume of flour originally provided.
- This gluten extraction is also referred to as “seitan” a meat substitute. Additional information regarding its use and recipes describing how it’s made are available online. You may also find pre-prepared seitan available for purchase in the grocery or health food store.
- As students are rinsing their cheesecloth bags, the starches are being solubilized and rinsing away.

Discussion

- Seitan is another meat substitute; can you think of another that uses the protein found in these field crops? (tofu)
- Both soy and wheat are two of the eight most common food allergens. All eight allergens are proteins and of all the macromolecules, it is proteins that are responsible for triggering immune responses in allergies. Having multiple alternative protein sources gives consumers options.
- Gluten is the rubber-like component of bread dough (it even sounds like glue, think of how you are able to knead bread dough). It is composed of two proteinaceous subunits, glutenin and gliadin.
- Gluten is insoluble in water. When you are rinsing with water, you wash the starches away and collect this hydrophobic protein.

***Experiment* – Protein analysis**

- Two possibilities are available for protein quantification, listed below. Choose one or both for use within this lab.
- Total protein content can be quantified by sending seed off to an analytical company (ex: Precision Soya or Bluegrass Farm). It is also possible that “average” data be presented to students, as opposed to repeating analytical work year after year.
- Qualitative protein analysis can be performed according to kit instructions for Bio-Rad Protein Analysis kit. If classroom is equipped with a spec-reader, this analysis can be quantitative.
- See “Are You Gonna Eat That?” curriculum on grownextgen.org for an explanation of both qualitative and quantitative protein analysis.

***Experiment* – Starch test**

- Starch is a complex carbohydrate
- Potassium iodide reagent (iodine-KI) or starch test papers (Precision Laboratories) can be used to test for starch.
- Using the semi-quantitative starch paper, have students solubilize 0.5 g of flour in 5 ml water. Have students dilute this initial solution 1:10 in series until each group has 5 unique solutions. [Transfer 1 ml of mixture of flour and water to 9 ml of water (diluting by a power of ten each time). Repeat this step until they have 5 total dilutions for each flour type.]
- Have students include a negative control, just water without flour.
- Following strip instructions, dip strip into solution for 1-2 sec. Incubate strip for 1 min before comparison to indicator chart. Repeat for each solution.
- Use the strip result with the highest dilution, which still registers on the color chart to determine the concentration. Be sure to account for each dilution step.
- Have students record results
- Note: Iodine in solution appears brown. The test is based on the principle that the iodide ions are able to form a complex within a starch helix. The formation of this complex results in the development of a blue color.

***Experiment* – Oil Extraction**

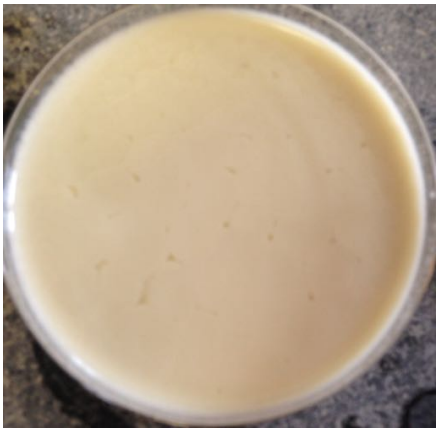
- For each seed, soak in 2x the water overnight.
- Macerate 5 g using a food processor or blender.
- Add 3 ml ethanol or acetone. Continue maceration.
- Decant off liquid into a secondary container (beaker, Petri dish, etc). Filter through a fine-gauged sieve or cheesecloth to remove particulate.
- Allow a small volume of liquid extractions to evaporate overnight on a wide, low container like half of a Petri dish
- Record approximate volumes of remaining oil. You can have students eyeball volume or use a micropipette to estimate. Some of the water soluble solids will still remain, qualitative analysis can easily be performed by assessing the “greasiness” of the residue.

Dry, cracked soybeans

Soaked overnight and drained

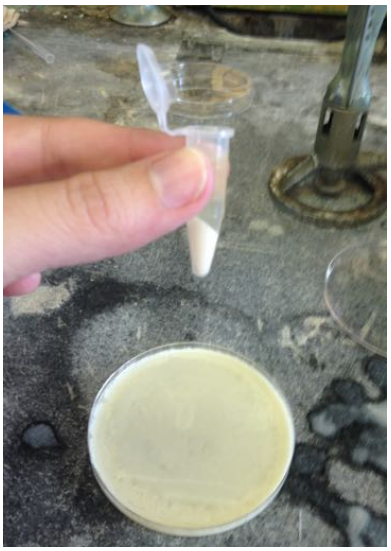
Milled in a food processor

Milled soybeans were mixed with 2x volumes acetone. After filtering twice through cheesecloth to remove the solids, soy extraction is shown below collected in a Petri dish. Hydrophobic fraction will soon start to separate (can be observed as cracks or veins in the dish below). This is one way we can initially determine which grains contain a high oil composition. To increase the portion of oil in the fraction extraction, students can use a pipet to specifically suck up and transfer the veins of hydrophobic liquid to the other half of the Petri dish lid.



Below is the remaining oil in the Petri dish after a brief drying period.

Alternatively, if teachers have access to a roto-vap or centrifuge, the solid fraction can be spun out of a liquid portion transferred to a microfuge tube.



After roto-vap for 2-3 hrs, or otherwise evaporating, remaining solids and hydrophobic fraction remains.

Discussion

-Have students evaluate their results for each of the qualitative and quantitative sections. What similarities do they notice among the results across field crops? What differences are observed/are some macromolecules found more abundantly in certain crops?