GROW
NEXT GEN

Photoperiod Experiment
Teacher Instructions

An important environmental factor to soybean growth and development is *photoperiod*, the number of hours of light a plant receives throughout the day. Species, even varieties, of plants are categorized as either short-day or long-day, based on how they flower in response to changes in photoperiod. While the category names used to describe each group use “day length”, it is actually the duration of the night that determines the photoperiod’s physiological response. In plants, bombardment by red light changes the structure and confirmation of the photoreceptive protein phytochrome, causing it to take on a biologically active shape. Inactive phytochrome is produced and found freely in the cytoplasm of the plant cell, but once it is activated, phytochrome is translocated and allowed to enter the cell’s nucleus, where it then acts with transcription factors to express genes that are photoperiod controlled.

Soybeans are short-day plants and, therefore, they flower earlier under short day lengths. For example, during the month of June day length in Columbus Ohio is about 15 hours, which is a long photoperiod and the month in which soybeans are in a vegetative developmental stage, whereas it is about 14 hours during the months of July and August when soybeans are actively flowering in their reproductive phase. In late September when the photoperiod is only 12 hours, soybeans are filling seeds and senescing; therefore, the time to harvest maturity is also photoperiod controlled. Because of this photoperiod sensitivity, soybean cultivars are classified according to their response to photoperiod. There are 8 maturity classes commonly used in the USA (0-VIII): in Ohio, the earliest maturing cultivars that are adapted to north of the state are designated II, and later maturing cultivars that are adapted to more southern locations are designated either III or IV. Maturity class increases when moving further south, with O being used in Minnesota and North Dakota to VIII in the Southern US.

In this lesson students will plant and care for soybeans under one of three different photoperiod regimes:

- 1st month-16 hours, 2nd month-14 hours, 3rd month 12 hours (control)
- 16 hours for all three months (long days)
- 12 hours for all three months (short days)

As the plants grow, students will analyze and maintain records of their plant’s progress. Students should also become familiar with the vegetative and reproductive stages of the soybean, more information and pictures of these stages can be found at the OSU crop extensions sites. Students should keep records of the dates in which they observe these reproductive transitions.

Prior to starting the activity and planting, students should be split up into groups of 3-4; each group will be responsible for a single 5-gallon pot of 3-4 mature soybeans at the end of the experiment. Teachers will need to have an adjustable timed growth light shelf for each photoperiod regime; if possible, the growth light from different regimes should not shine on one another. It is recommended that the growing shelves have light fixtures totaling at the least 6 fluorescent bulbs (the more lights the better), and that the fixtures be suspended in a way that allows for light height adjustment as the plants grow taller (shop light fixtures work well). Use soil made for containers. Do not use pure garden soil, because it will clump and will not drain properly. Soybeans need container mix that will drain well. A standard container soil should suffice. Make your own container mix by combining equal parts peat moss, perlite and garden soil. The initial planting will take a full 60 minutes; students should subsequently make observations for 10 to 15 minutes twice a week until tissues and seeds have been harvested.

Day 1:
Activity – Planting

- In large plastic bins, soak standard potting mix using water until slightly moist. Fill pots with slightly moistened growing media to about 3/4s up to soil line
- Spread half a cup of slow release fertilizer evenly on top
- Continue to fill the pots to the soil line with slightly moistened soil
- Poke 8 one inch deep hole in the top of the growing media using your finger
- Place a soybean in each whole and light cover with dry growing media
- Using a pipette or dropper, gently apply water to each seed (10-20 drops)
- Label your groups pot properly and place it on the proper lighted shelf
- Make sure light timers are programmed to their proper photoperiods

Growing Tips

- Adjust lights as the plants grow so that the lights are 10-15 cm from the growing meristem
- Keep room temperature above 60°F
- Check on plant hydration everyday and water as needed
  - Plants will need watered more often as they grow in size
  - If you lift the pot and it feels lite, it may need to be watered
- Thin to 3-4 plants per pot; transplant from other pots if necessary
  - Thin and transplant as soon as you are confident in the establishment of the seedlings
- Coinciding grow lights with times there are other lights sources helps to prevent light contamination

Discussion

- Prior to planting, give students an overview of the experiment.
- Explain to the students what the control and experimental groups are and that the two groups are the same except for one variable (photoperiod)

Days 2-7: Germination

- Have students make observations and record the dates of germination
    - Include germination rates and note any unusual features
    - Do the germination rates and dates differ between the groups?
- After the first week, seedlings will appear. Seedlings should be thinned using scissors or forceps so that only 3-4 plants remain per pot
    - If needed, transplant seedlings from crowded pots and plant them in pots that need them
- The seed coat may not fully fall off the emerging seedling, causing the cotyledons to not fully expand. The remainder of the seed coat should be removed carefully with forceps

Discussion Points

- Germination starts with and seed and ends with a fully expanded seedling
- Each seed contains a single embryo and enough stored energy/nutrients to give the embryo a head-start in its growth
- Germination is initiated when the seed **imbibes** water from the growing media (why we need to use moist soil); this intake of water causes the dried and dormant tissues of the seed to swell and crack the outside layer of the seed, **seed coat**
- An embryonic root, the **radicle**, will soon emerge and grow downwards
    - The radicle soon develops fine root hairs that greatly increase the roots surface area; in turn, increasing the water uptake of the whole plant
- The new uptake of water causes the cell of the embryonic stem, **hypocotyl**, to swell and elongate, causing the seed to become upright and emerge from the soil
- As the seedling emerges and casts off the seed coat, two embryonic leaves, **cotyledons**, fully expand and begin to produce chlorophyll.
    - The cotyledons will begin photosynthesizing, but will look different than the future leaves. That is because the cotyledon's primarily act as storage for essential nutrients and energy the seedling needs to quickly grow
- While all **angiosperms**, flowering plants, germinate from seeds, they don't all follow the previously outline steps.
    - For example, soybeans are **dicots** and germinate with two cotyledons; while corn and rice, being **monocots**, germinate with one cotyledon.
- Despite having a great understanding of germination in higher plants, scientist continue to have question and perform research to better understand the mechanisms and environmental factors that control germination

Days 10-30: Growth and Development

- Weekly, have students make observations and record the data on the progress of their plants
    - Measure and/or record the following for each plant:
        - Number of leaves
        - Plant height from soil to **apical meristem**, growing point
        - Number of leaf attachment areas, **nodes**
        - Date of first flower
        - Total number of flowers
        - Number of flowering nodes
    - Items and changes to notice:
        - Leaf shape, **morphology**, will change as plants grow
        - Length of stem segments between leaves, **internodes**
        - Developing expansion of new leaves
- By day 30, there should be visible differences between photoperiod regimes
- The data collected throughout the weeks will help students quantify these changes
- Students should also become familiar with the vegetative and reproductive stages of the soybean, more information and pictures of these stages can be found at the OSU crop extensions sites.
- As these stages come about, their dates of occurrence should be recorded
- As plants grow, they will require staking

Discussion Points

- As the plant matures, it undergoes **growth**, the additions of new cells and elongation of cells, and **development**, the arise of different tissue and organs
- As the soybean grows, it adds new nodes from the apical meristem and elongates internodes, both causing the plant to increase in height
    - New leaves and nodes always arrive from a meristem
    - Each new organ emerges and overtime grows larger
- Plants may also start to form branches at lower nodes as the apical meristem gets further from the lower nodes
    - This branching is caused by the gaining and losing of plant **hormones**, chemical signals, from the apical meristem
- Growth allows plants to “reach” new areas for photosynthesis and place their leaves and flowers in positions that are conducive to their purpose
- Leaves produce carbohydrates by combining carbon dioxide and water in the presence of light, this process is called **photosynthesis**
- These sugars are then transported throughout the plant via the stem; simultaneously the stem also transports water from the roots to the leaves
- The emergence of flowers denotes a change from the plants vegetative stage to a reproductive one
    - From then on, the plant is solely focused on creating more progeny and producing healthy seeds

Days 30-45: Flowers and Pollination

- Weekly, have students make observations and record the data on the progress of their plants
    - Measure and/or record the following for each plant:
        - Number of nodes with flowers
        - Plant height from soil to apical meristem
        - Total number of flowers
        - Number of flowering nodes
    - Items and changes to notice:
        - Leaf shape, **morphology**, will change as plants grow
        - Length of stem segments between leaves, **internodes**
        - Developing expansion of new leaves
- By day 30, there should be visible differences between photoperiod regimes
- The data collected throughout the weeks will help students quantify these changes

Days 45-90: Fertilization and Seed Development

- Weekly, have students make observations and record the data on the progress of their plants
    - Measure and/or record the following for each plant:
        - Number of leaves
        - Plant height from soil to **apical meristem**, growing point
        - Number of leaf attachment areas, **nodes**
        - Date of first flower
        - Total number of flowers
        - Number of flowering nodes
    - Items and changes to notice:
        - Leaf shape, **morphology**, will change as plants grow
        - Length of stem segments between leaves, **internodes**
        - Developing expansion of new leaves
- By day 30, there should be visible differences between photoperiod regimes
- The data collected throughout the weeks will help students quantify changes
- Instructors may choose to end the experiment at any point after 45 days
    - It is advised to wait till there is a visible significance difference between the photoperiod regimes
    - Plants can be grown through maturity and all the way to harvestable dry seeds, or the experiment can be finished after pod formation and embryos are counted
        - The longer an instructor waits the more data there is to collect, but this additional time also allows for problems to arise (plants dying early, pest build up, etc.)
- After data collection, instructors can compile data from all groups and present the total data to the class
    - Look at which traits and characteristics have the most variability between and within the different photoperiod regimes