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LIGHT AND PLANTS

**A Series of Experiments
Demonstrating Light Effects
On Seed Germination, Plant
Growth, and Plant Development**

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**Agricultural Research Service
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LIGHT AND PLANTS

A Series of Experiments Demonstrating Light Effects on Seed Germination, Plant Growth, and Plant Development

By R. J. Downs, H. A. Borthwick, and A. A. Piringer¹

INTRODUCTION

Each year scientists in the U. S. Department of Agriculture receive many inquiries from students, teachers, and other interested biologists for details of simple but dramatic experiments to demonstrate the photomorphogenic effects of light on plants. To answer these requests for this specialized information detailed and systematic experiments and demonstrations on effects of light on seed germination, growth, flowering, and fruiting are outlined herein. References accompanying each experiment are cited for supplementary reading and additional details. Certain of these references will not be readily available to all interested persons, but, in general, the cited papers can be obtained from college and other school libraries of most metropolitan areas as well as the personal libraries of local plant scientists.

LIGHT AND SEED GERMINATION

Seeds of many kinds of plants germinate poorly or not at all when planted and covered with soil. In many instances these are seeds that require light for germination. Some seeds, such as those of peppergrass (Lepid-

ium virginicum), do not germinate at all in darkness. Others, such as seeds of Grand Rapids lettuce (Lactuca sativa), often germinate as much as 30 percent in darkness and some lots even higher. However, all the seeds of both peppergrass and Grand Rapids lettuce germinate following a single brief exposure to light. A single exposure to light, nevertheless, is not adequate to promote germination of all kinds of light-sensitive seeds. Seeds of the Empress tree (Paulownia tomentosa), for example, require long periods of light for several days.

Germination of still other kinds of seeds, such as those of henbit (Lamium amplexicaule), appears to be inhibited by light.

A favorable temperature is one of the requirements for germination and often a change or alternation of temperatures is more effective than a constant one in securing maximum germination. For example, only about 30 percent of peppergrass seeds imbibed in water and placed on blotters in Petri dishes at a constant temperature of 70° F. may germinate in the light. If the temperature is alternated, more seeds germinate. If the seeds are imbibed in a solution

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containing 0.02-percent potassium nitrate and the temperature alternated, maximum germination is attained. Such an alternation of temperatures might be 77° F. for 8 hours per day and 60° F. for 16 hours per day.

Germination studies are often made on blotters in Petri dishes in order to facilitate handling, planting, and counting the seeds. If Petri dishes are not available, plastic sandwich boxes with lids, and filter paper or paper towels will perform the same job. Studies of the effect of light on germination of seeds implies that some of the seeds must be kept in the dark to act as a check or control. The term "dark" means "total darkness," a complete absence of light. To provide the darkness required for the dark controls a proven method is to make bags of at least two layers of black sateen cloth. These bags must be large enough to contain the dishes, with enough slack at the opening so that a flap may be folded back to prevent entrance of light. An alternate method would be to cover the dishes with two or more layers of aluminum foil.

Studies on the effect of light on various plant responses can be made in greater detail using red and far-red² radiant energy. These wavelengths are the most effective ones for regulating many plant responses to light and they can be obtained by using colored filters in conjunction with the proper light source. The fluorescent lamp emits considerable red but almost no far-red light and is, therefore, used as a source of red light. A filter of two layers of red cellophane removes all visible light except red and since very little far-red is emitted by the lamp, the net result is reasonably pure red light.

Incandescent-filament lamps emit considerable amounts of far-red and

are thus good sources of far red. The visible light is removed by appropriate filters such as a combination of red and blue cellophane. The red cellophane absorbs all the visible light except red and the dark-blue cellophane absorbs red. However, neither color of cellophane absorbs far red, so the radiation passing through the filter is, therefore, far-red.

Seeds of trees, shrubs, ornamentals, vegetables, grains, and grasses can be obtained from commercial seed sources that range from special seed supply houses to the local hardware store. Many weed seeds are light-sensitive and these can be gathered by the investigator. After gathering the seeds they should be stored dry in a refrigerator (about 40° F.) until an appropriate time to begin the experiments because at higher temperatures they often undergo change in their light requirements. A good supply of seeds should be gathered to assure an adequate amount for possible additional experiments. Any difficulty in identifying the plants can be resolved with the aid of high school biology teachers, the botanists or horticulturists at the State University, State Agricultural Experiment Stations, or agriculture extension specialists.

Demonstrations A-1 through A-5 tell us the following facts:

- Certain kinds of seeds require light in order to germinate.

- The light requirement is not something that occurs only under a special set of experimental conditions, but occurs when seeds are planted in an ordinary way in pots of soil.

- A light requirement can be induced in seeds that normally do not require light for germination.

² In Europe this would be referred to as near infrared.

- The photoreaction that allows germination to proceed is reversible; red radiant energy drives the reaction in one direction and far-red drives it in the reverse direction.

- The sensitivity of seeds to a given amount of radiant energy changes with the period of imbibition.

Several questions should come to mind immediately. How much light is required to induce germination? What are the relative amounts of red and far-red required to drive the reactions? Do all light-sensitive seeds require the same amount of energy to trigger germination? What is the effect of various temperatures on the light requirements? Are there other methods of inducing a light requirement in seeds that normally are not light-sensitive? Are there any seeds that are inhibited from germinating by light? Experiments can be designed to answer these and many more questions relating to the mechanism by which light controls germination.

LIGHT AND PLANT GROWTH

Vegetative growth of plants is to a large degree controlled by light. Plants grown in total darkness have very long internodes, small leaves, and are yellow in color because no chlorophyll is formed. If the dark-grown plants are exposed to weak light for a minute or two each day, the plants have shorter internodes and normal-size leaves, although they may still be yellow and without visible chlorophyll. Daily exposures of the plants to light of higher intensities or for a longer duration may not change the size of the leaves or internodes of the plants from that obtained with brief exposures to light of low intensity, but the plants turn green as chlorophyll is formed.

The formative effects of light, but not chlorophyll formation, result from the same red, far-red reversible photoreaction that also controls flowering of photoperiodically-sensitive plants, germination of light-sensitive seeds, and many other plant responses. Red is the most efficient portion of the spectrum in inhibiting stem elongation and promoting leaf expansion. A far-red irradiation immediately following the red reverses the potential effect of the red irradiation and the stems become long.

If light is directed at either light-grown or dark-grown plants from one side, the leaves tend to bend and the leaf petioles twist until the plane of the leaf blade is perpendicular to the light. The stems tend to curve in such a way that the tip of the stem is directed toward the light source. This phenomenon is called "phototropism" and is caused by a different photoreaction than the red, far-red one. Blue light is the most effective kind of light to promote the phototropic response.

Demonstrations B-1 through B-4 show several ways in which light influences plant growth and development. These demonstrations tell us the following facts:

- Light inhibits stem growth and promotes leaf expansion.
- Plants bend toward the light.
- Chlorophyll formation requires light and the light must be of higher intensity than that which controls stem length.
- The red, far-red reversible photoreaction that controls seed germination also controls stem length and leaf size.

Additional experiments can be designed to answer many other questions relating to the manner by which light controls plant growth. Examples of

such questions are as follows: Are bending (phototropism) and growth of internodes controlled by the same photoreaction? This question can be answered by using different regions of the spectrum (colors of light) and testing to see if bending and growth are controlled by the same colors.

Does the duration of darkness following the far-red irradiation of light-grown plants affect the ultimate length of the internodes? What is the optimum period of darkness and why is it optimum?

How concentrated is the pigment that controls growth? How do we know it is not chlorophyll? These questions can be answered by comparing the growth responses of albino and green corn or barley seedlings.

LIGHT AND PLANT PIGMENTS

The autumn coloration of leaves and stems of woody plants is in part due to the formation of a red pigment called anthocyanin. The formation of anthocyanin is also responsible for the red color of apple fruits and for the red to purple color of milo, turnip, and cabbage seedlings.

A common observation is that apples often do not turn red uniformly but that one side of the fruit is green or at least a lighter shade of red than the other side. The reddest side of the apple is usually facing outward from the tree. The formation of the red color (anthocyanin) in apple fruits is, of course, controlled by light. Detailed studies have shown that anthocyanin formation in milo, turnip, and cabbage seedlings and in leaves of red maple and other trees is also regulated by light.

Unlike many other light-controlled plant responses anthocyanin formation requires high intensity light for a relatively long time. However, at the close of the high-intensity light period

the low intensity, red, far-red photoreaction may exert final control on anthocyanin synthesis. Thus, if the plant material is irradiated for a few minutes with far-red at the close of the high-intensity light period, the potential anthocyanin synthesis is inhibited and very little is formed. If a brief irradiation with red follows the far-red, then anthocyanin is formed in an amount equal to that produced by the high-intensity light alone.

An example of a low-intensity, light-controlled coloration is the yellow color of the skin of the tomato fruit. Plant breeders recognize differences in the color of the skins of fruits of certain tomato varieties and have classified the skins as yellow or clear. The red flesh and a transparent or white skin give the fruit a translucent pink color, whereas the yellow skin and red flesh give the fruit an orange-red appearance. In many tomato varieties the formation of this yellow pigment is controlled by light. Moreover, the same red, far-red photoreaction that controls flowering of photoperiodically-sensitive plants, germination of light-sensitive seeds, and many other plant responses also controls the formation of the yellow pigment in the skins of tomato fruit.

Demonstrations C-1 through C-4 concern light and its control of plant coloration. From these demonstrations we know:

- That light is required for the formation of the red color (anthocyanin) of certain seedlings and apple fruits.
- Light is required for the formation of a yellow pigment in the skin of tomato fruit.
- The coloration occurs only in the areas that received light--there is no translocation of the stimulus.

Additional experiments can be designed to learn more about the light reaction and about the chemical processes that result in pigment formation. Questions that one might ask are: How much energy is required to induce the formation of anthocyanin? As light energy is increased, does the amount of anthocyanin increase proportionately? Once the light requirement is fulfilled, what is the rate of anthocyanin formation? What is the role of temperature? What is the role of sugar? Does the red, far-red reversible photoreaction operate in the control of coloration?

EFFECT OF DURATION OF LIGHT ON PLANTS

Flowering of many kinds of plants is controlled by the relative length of the daily light and dark periods. This phenomenon is called "photoperiodism." Some plants, such as certain varieties of chrysanthemum, poinsettia, morning-glory, cocklebur, and lamb's-quarter, are short-day plants and flower only when the days are short and the nights are long. Certain varieties of spinach, beet, barley, and tuberous-rooted begonia, are examples of long-day plants which flower only when the days are long and the nights are short. Flowering of many other kinds of plants is hastened but not absolutely controlled by the appropriate daylength. For example, scarlet sage, variety America, flowers quickly on short days but eventually flowers on long ones. Many varieties of petunia flower most rapidly on long days but finally flower on daylengths as short as 8 hours.

Bulbing and tuber formation are also controlled by daylength. Tuberous-rooted begonia, which is a long-day plant for flowering, produces tubers on short days but not on long days. Onions, on the other hand, produce bulbs on long days but not when the days are short.

Dormancy and thereby preparation of woody plants for the coming of winter is another plant response regulated by photoperiod. Even in the warm greenhouse many woody plants stop elongation of stems, produce terminal buds, and "harden off" when the days begin to shorten in the autumn. However, if artificial light is used to keep the days long, plants in the warm greenhouse will continue growing during the naturally short days of winter and several years' "field" growth is often obtained in only 1 year.

These plant responses are regulated not by the length of the light period but by the length of the dark period. Thus, a long-day plant is really a short-night plant, and a short-day plant is really a long-night plant. Therefore, a long-day plant will flower, dormancy will be prevented, and onions will produce bulbs when a long dark period is broken into two short periods by a relatively brief exposure to light near the middle of the dark period. Under these same conditions, short-day plants will remain vegetative.

Studies of the responses of green plants often require plants to be grown indoors, yet they should have the same healthy appearance as well-tended plants grown out-of-doors. Everyone knows that plants cannot survive without light of adequate intensity to operate the processes of photosynthesis. In the field and garden or in the greenhouse this high-intensity light is obtained from the sun which often provides an illumination as high as 10,000 footcandles. In the average home the light intensity is usually too low for growth of many kinds of plants, even on the window sills. However, plants can be grown quite successfully with artificial light in complete absence of sunlight. Beans, tomato, cereals, and many ornamentals that grow in open sunlight make satisfactory growth if the

artificial light intensity is about 1,000 footcandles. Shade-loving plants, such as African violets, begonias, episcias, gloxinias, and orchids, will grow well with intensities as low as 500 footcandles.

A practical source of artificial light for plant growth is the fluorescent lamp. These lamps supply the necessary intensity without excessive heat and are available in various lengths, wattages, and colors. They are usually operated on one- or two-lamp ballasts, which maintain the proper current and provide the starting voltage. Prewired lamps and ballasts of several sizes and types are available as commercial luminaires or as channels.

Many kinds of plants can be grown satisfactorily using only two 40-watt fluorescent lamps. Since the lamps themselves are relatively cool, the plants may be placed quite close to them without danger of excessive heat or burning. Table 1 shows the illumination at various distances from two 40-watt cool-white fluorescent lamps mounted 2 inches apart. If the lamps are mounted further apart, the illumination at 6 inches or less from the lamps is markedly decreased.

If the daylength is to be controlled, plants must be put into complete darkness at the close of a particular photoperiod. A dark chamber can be made of masonite or plywood with caulked seams, or it could be made of two or more thicknesses of black sateen cloth stretched over a wooden frame. If used carefully, a cardboard box with all seams and joints sealed with paper tape could be placed over the plants to provide darkness.

Experimental procedures can be facilitated and made more exact if an electric time switch is available to turn the lights on and off at any desired time.

Table 1.--Illumination in footcandles at various distances from two or four 40-watt standard cool-white fluorescent lamps mounted approximately 2 inches from a white-painted reflecting surface.

Distance from lamps	Illumination		
	Two lamps*	Four lamps*	
	Used**	Used**	New
(in)	(fc)	(fc)	(fc)
1	1,100	1,600	1,800
2	860	1,400	1,600
3	680	1,300	1,400
4	570	1,100	1,300
5	500	940	1,150
6	420	820	1,000
7	360	720	900
8	330	660	830
9	300	600	780
10	280	560	720
11	260	510	660
12	240	480	600
18	130	320	420
24	100	190	260

* Center-to-center distance between the lamps was 2 inches.

** These lamps had been used for approximately 200 hours.

Demonstrations D-1 through D-4 show some of the effects of the relative lengths of day and night on plant growth and reproduction. These demonstrations tell us that:

- Some plants flower on short days and long nights, whereas others flower on long days and short nights.
- Dormancy of woody plants in the autumn is brought about by short days.
- Daylength controls tuber and bulb formation as well as flowering and dormancy.

Additional experiments can be designed to answer and demonstrate many other aspects of the photo-periodic control of flowering bulbing, and dormancy. For example, we might ask, what is the critical daylength for short-day plants? What is the longest day (shortest night) that will include flowering in short-day plants? What is the shortest day (longest night) that will induce flowering of long-day plants? When a long dark period is interrupted by a brief interval of light, what is the minimum energy required to keep short-day plants vegetative or to induce flowering of long-day plants? When is the most efficient time to give the interruption during the dark period? Is the control of flowering operated through the same red, far-red reversible photoreaction that controls other plant responses?

GENERAL CULTURAL HINTS

For all demonstrations in which seeds are germinated in soil, or in which young seedlings are to be grown, the soil should be sterilized. Sterilizing the soil destroys harmful insects, disease-producing organisms, and weed seeds. Soil may be sterilized by different methods as follows: (a) Place small lots of moist soil in a shallow pan and bake for at least 1 hour at a temperature of 215° F., then cool but do not use until at least 2 weeks; (b) place soil in an autoclave or pressure cooker and steam sterilize at 15 pounds' pressure for at least 1/2 hour, then allow to stand for a minimum of 2 weeks; (c) sprinkle 1 quart of formaldehyde solution (1 pint 37 percent commercial formaldehyde to 3-3/4 gallons water) on 1 square foot by 6 inches of soil placed in a box or bushel basket, then water liberally and completely cover with

plastic or heavy cloth for 48 hours, stirring frequently to hasten escape of the formaldehyde gas, and allow 2 weeks before use of the soil (CAUTION: Do not use for planting as long as fumes are present, because formaldehyde gas is an irritating poison to humans and is toxic to plants.)

Plants are usually grown in clay pots of 3-, 3-1/2-, or 4-inch diameter filled with sterilized soil. Before the soil is put into the pot a piece of broken pot is placed in the bottom to cover the hole so that the soil will not plug it and prevent good drainage. Clean pots should always be used.

When pots are not available or are for some reason objectionable, plastic cups, polyethylene freezer food containers, or even tin cans may be used. One or more holes should be punched in the bottom of these containers and the holes covered with fiber-glass matting or plastic window screen before filling with soil. Good drainage is imperative for good plant growth.

Studies of the effect of light on plant growth and flowering require that the plants be placed in darkness at certain times. As with seed germination, this means complete or total darkness. Because plants require more space than seeds, light-tight bags are usually not satisfactory. Instead, a dark chamber must be constructed in such a way that there is adequate air exchange between the inside and outside of the chamber to prevent overheating. A satisfactory and proven method is to construct a frame of wood and cover it with at least two layers of black sateen cloth. An entrance or door can be provided by making an overlapping flap.

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DEMONSTRATIONS

A. Light and Seed Germination

DEMONSTRATION A-1: Effect of light on germination of seeds planted in soil.

Materials:

1. Seeds of peppergrass (Lepidium virginicum or L. densiflorum).
2. Sterilized potting soil.
3. Six clay pots, or other suitable containers, 3 to 4 inches in diameter.
4. Small glass squares large enough to completely cover the tops of the pots.
5. Glass baking dish or enamel pan large enough to contain all the pots.
6. Pot labels.

Procedure:

1. Fill six pots or containers with moist (not wet) sterilized soil to within 2 centimeters of the top of the pots. Smooth the soil surface and tamp the soil gently but firmly with the bottom of one of the pots.
2. Prepare six lots of 100 seeds each and distribute one lot of seeds uniformly over the surface of the soil in each pot. Treat the pots as follows:
 - (a) Pot 1 - leave the seeds on the surface. Do not cover them with soil.
 - (b) Pots 2 to 6 - cover the seeds with 1 centimeter of soil. Level the soil surface and tamp gently.
3. Do not water the top surface of the soil. Place all the pots in the large glass dish or enameled pan and sub-irrigate the soil in the pots by adding water to the dish. Maintain the pots in this dish, being careful to have them setting in about 1 centimeter of water at all times.
4. Place all pots in the light.
5. Keep each pot covered with a glass square, at least until the seedlings that will develop are well established. The soil will be kept moist by capillary action. The glass cover will admit light but will prevent excessive water loss from the soil and maintain a high humidity at the soil surface. This is important during the critical periods of germination and early seedling growth.
6. Write the name of the plant material, the date of planting, the date of treatment on a label and insert it into the soil at the edge of the pot.
7. Treatments:
 - (a) Pot 1 - see 2a.
 - (b) Pot 2 - see 2b.
 - (c) Pot 3 - immediately after covering the seeds with soil make a narrow slit 3 to 4 centimeters deep in the soil across the diameter of the pot with a knife.
 - (d) Pots 4, 5, and 6 - repeat the process described for pot 3, 1, 2, and 4 weeks after planting, respectively.

Observations:

Record the date of exposure to light, the subsequent date of germination and the extent of germination. Moist peppergrass seeds exposed to light (as in pot 1) will germinate in 3 to 4 days after exposure. Seeds covered with 1 centimeter of soil will not germinate since they are in the dark (as in pot 2). Slitting the soil with a knife blade exposes some of the buried seeds to light. Thus (as in pots 3 to 6) germination of seeds occur in the slit made in the soil.

The soil may shrink away from the sides of the pot and expose some seeds to light and seedlings may appear. This can be avoided by planting the seeds away from the edge of the pots. Slitting the soil at regular intervals after planting illustrates that germination will occur any time the seeds are exposed to light. Slitting the soil, in effect, simulates field cultivation. Thus, cultivation, while destroying plants and seedlings, also brings weed seeds such as peppergrass to the surface of the soil, where they receive light, germinate, and produce more weeds.

Supplementary Reading:

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DEMONSTRATION A-2: How to test various kinds of seeds to determine their light requirement for germination.

Materials:

1. A minimum of four Petri dishes or plastic sandwich boxes with lids.
2. Ordinary color-fast or white blotters, filter paper, or paper towels.
3. Black sateen cloth bags made of two layers of cloth large enough to hold the dishes. An alternate method is to wrap the dishes in two layers of aluminum foil.
4. Seeds of several kinds of weeds. (Although some kinds of seeds are known to be light-sensitive, many kinds have never been tested. This is especially true for weed seeds, so they would be the more interesting group to investigate.)

Procedure:

1. Collect seeds of several kinds of local weeds. In general, seeds will retain their viability fairly well when stored dry in a refrigerator. Some suggested seeds known to be light-sensitive are peppergrass (Lepidium virginicum and L. densiflorum), henbit (Lamium amplexicaule), and

hedge mustard (Sisymbrium officinale). Other weed seeds worthy of investigation are Shepard's purse (Capsella bursapastoris), yellow rocket (Barbarea vulgaris), tumble-mustard (Sisymbrium altissimum), chickweed (Stellaria ssp. and Cerastium ssp.), sheep sorrel (Rumex acetosella), the small-seeded cacti, and, of course, many others.

2. Four dishes should be used for each kind of seeds tested.
3. Cut the blotters to fit the dishes and presoak overnight (about 16 hours) by putting enough tap water into the dishes to flood the blotters.
4. After the blotters are thoroughly soaked, pour off the excess water and evenly distribute 100 seeds over the surface of the blotters in each dish.
5. Immediately after the seeds are distributed, place the covers on the dishes and place them in the black cloth bags.
6. Allow the seeds to imbibe water in the dark for a period of 16 to 24 hours, then begin treatments.
7. Treatments:
 - (a) Dishes 1 and 2 should be kept at about 70° F. during the entire period of the demonstration. Dishes 3 and 4 should be held at about 60° F. during the 16- to 24-hour imbibition period, then transferred to a temperature of 77° F. for the remainder of the demonstration.
 - (b) Dishes 1 and 3 are placed in the black cloth bags at the time of planting and left there throughout the demonstration. These will serve as "dark controls."
 - (c) Dishes 2 and 4 should be placed in the light for 1 hour each day. The temperature during the period of irradiation can be between 70° and 80° F. Light from two 40-watt standard cool-white fluorescent lamps should be adequate.
 - (d) When the seeds in dishes 2 and 4 have germinated, the other dishes are removed from the black cloth bags and the number of germinated seeds counted and recorded for each treatment.

Observations:

Record the number of days required for germination, the temperature, light conditions, and so forth. Count the number of seeds that germinate under each treatment and record as percent germination. When a light-sensitive seed is found, demonstrations can be designed to determine how much light the seeds require, how many times they must be exposed to light, and such. These seeds can also be used in Demonstrations 3, 4, and 5.

Supplementary Reading:

See Demonstration A-1.

DEMONSTRATION A-3: Effect of duration of imbibition (soaking) period on effectiveness of a given light exposure in promoting germination of light-sensitive seeds.

Materials:

1. Eight Petri dishes or plastic sandwich boxes with lids.
2. Two to four thicknesses of blotters, filter paper, or paper towel cut to fit the dishes.

3. Eight black sateen cloth bags made of two layers of cloth large enough to hold the dishes. As an alternate method dishes can be placed between the folds of a large, double layer of black cloth, or they can be wrapped with two layers of aluminum foil.
4. Light-sensitive seeds such as Grand Rapids lettuce or peppergrass (Lepidium virginicum).

Procedure:

1. Prepare the dishes as outlined in the procedure in Demonstration A-2. Use 0.2 percent KNO_3 instead of tap water for peppergrass.
2. All dishes except dish 2 are immediately placed in darkness (in the black cloth bags or between folds of a black cloth "blanket.") Dish 2 is exposed to light for a period of 5 minutes, then placed in darkness. Illumination provided by two 40-watt fluorescent lamps is adequate.
3. Treatments:
 - (a) Dish 1 remains in darkness throughout the demonstration and serves as the dark control.
 - (b) Dish 2 is irradiated immediately after distributing the seeds, then placed in darkness.
 - (c) Dish 3 is irradiated for a period of 5 minutes with light from the fluorescent lamps after the seeds have imbibed in darkness for a period of 1 hour; that is, the seeds are exposed to light 1 hour after soaking.
 - (d) Dishes 4, 5, 6, 7, and 8 are exposed to 5 minutes of light after 2, 4, 8, 16, and 24 hours of imbibition in darkness.
4. Following the 5-minute exposure to the fluorescent light the dishes are returned to the black cloth bags, to the folded black cloth blanket, or to the aluminum foil.
5. Four days after the seeds were "planted" the dishes can be removed from the dark and the number of germinated seeds counted and recorded.

Observations:

Count the number of seeds germinated in each dish and record as percent germination. These data can be presented in a line graph by plotting percent germination against the number of hours of imbibition. The results may show the sensitivity of the seeds to a given dose of light changes during the period of imbibition.

Supplementary Reading:

See Demonstration A-1.

DEMONSTRATION A-4: How a light requirement for germination can be induced in seeds that normally do not require light for germination.

Materials:

1. Petri dishes or plastic sandwich boxes with lids.
2. Blotters, filter paper, or paper towels cut to fit the dishes.

3. Black sateen cloth bags made of two layers of cloth large enough to hold at least two dishes.
4. Dark-blue cellophane.
5. Fluorescent lamp (a fluorescent desk lamp will do).
6. Seeds of several kinds of plants including several varieties of lettuce and tomato.

Procedure:

1. Prepare dishes as previously described in Demonstration A-2.
2. Distribute 100 seeds evenly on the blotters of each dish.
3. Dish 1 is placed in a black cloth bag (darkness).
4. Dish 2 is placed under the fluorescent lamp.
5. Dishes 3 and 4 are completely covered with two layers of dark-blue cellophane and placed under the fluorescent lamp.
6. The fluorescent lamp is left on continuously.
7. After germination is apparent in dish 2, count and record the number of germinated seeds in all dishes.
8. Re-cover dish 3 with the blue cellophane and replace both dishes 3 and 4 (without cellophane) under the fluorescent lamp.
9. When germination is completed in dish 4, count and record the germinated seeds in both dishes 3 and 4.

Observations:

The seeds in the dishes under the dark-blue cellophane may not germinate, whereas those receiving either light or darkness may germinate nearly 100 percent. After a dish has remained under blue cellophane in light for 3 or 4 days it can then be covered with black cloth and the seeds may remain dormant in the dark. When subsequently given unfiltered light, they promptly germinate.

Supplementary Reading:

See Demonstration A-1.

DEMONSTRATION A-5: Photoreversible control of seed germination by red and far-red light.

Materials:

1. Three Petri dishes or plastic sandwich boxes with lids.
2. Ordinary color-fast or white blotters, filter paper, or paper towels.
3. Red and dark-blue cellophane.
4. Black sateen cloth bags made of two layers of cloth large enough to hold each dish.
5. Light-sensitive seeds such as Grand Rapids lettuce or peppergrass (Lepidium virginicum).

Procedure:

1. Cut two to four layers of blotter to fit each dish and presoak overnight (about 16 hours) by putting enough tap water into the dishes to flood the blotters.
2. After the blotters are thoroughly soaked pour off excess water and evenly distribute 100 seeds over the surface of the blotters in each dish.
3. Immediately place the dishes with lids in the black cloth bags.
4. Allow the seeds to imbibe water in the darkness of the black cloth bags for a period of 16 to 24 hours, then begin treatments.
5. Treatments:
 - (a) In the dimmest light possible, preferably complete darkness, remove dishes 1 and 2 from their black cloth bags and wrap each dish with two layers of red cellophane.
 - (b) Place both the cellophane-wrapped dishes under the fluorescent lights for a period of 5 minutes.
 - (c) Return dish 1 to its black cloth bag without further exposure to light. If no dark room is available during this transfer, place the dish in the black cloth bag without removing it from the red-cellophane wrapping.
 - (d) Dish 2 is wrapped in blue cellophane so that the seeds are now covered with two layers of red and two layers of blue cellophane.
 - (e) Dish 2 is now exposed to light from the incandescent lamps for a period of 15 minutes.
 - (f) Place dish 2 in the black cloth, either in complete darkness or if necessary still enclosed in the red- and blue-cellophane wrapping.
6. The three dishes of seeds have now received their treatments. Dish 3 has remained in the black cloth bag and serves as a dark control. Dish 1 has been exposed to red radiant energy for 5 minutes and dish 2 has been exposed to red for 5 minutes and to far-red for 15 minutes.
7. Allow 3 to 4 days to elapse, then remove the dishes from their black cloth bags and count and record the number of germinated seeds. Temperatures should be held as close to 70° F. as possible.

Observations:

When counting the number of seeds germinated in each dish, record as percent germination. These data can be presented in either tabular form or in a bar graph, using the bars for treatments and the height of the bars as percent germination. Those seeds that remained in darkness will probably germinate 0 percent if peppergrass seeds were used, or 5 to 25 percent if seeds of Grand Rapids lettuce were used. Those seeds receiving red light will probably germinate 90 to 100 percent for both species, whereas those receiving red followed by far-red might germinate 5 to 10 percent for peppergrass, and 5 to 25 percent for lettuce. Evidence has now been obtained to show that these seeds require light (red) for germination, and that the potential germination induced by the exposure to red can be reversed by a subsequent exposure to far-red radiant energy.

Supplementary Reading:

See Demonstration A-1.

B. Light and Plant Growth

DEMONSTRATION B-1: Control by light of the growth of an internode.

Materials:

1. Corn seeds.
2. Two 4- to 5-inch clay pots or other suitable containers.
3. Sterilized soil.

Procedure:

1. Place a piece of broken pot, fiber-glass mat, or plastic screen over the drainage hole in the bottom of the pot or container.
2. Fill pot or container 1 with sterilized soil to within 3 centimeters of the top of the pot, tamp the soil gently, place 3 to 4 corn seeds on the surface of the soil, and cover them with 2 centimeters of soil. Tamp firmly.
3. Fill pot 2 with 3 centimeters of sterilized soil, tamp gently, place 3 to 4 corn seeds on the surface of the soil, and fill the pot with sufficient soil to reach the same level as in pot 1. Tamp firmly.
4. Place both pots in a large glass dish or enameled pan and sub-irrigate by adding water to the dish or pan.
5. Place both pots in the light at a temperature of about 70° to 80° F.
6. The seedlings of pot 1 will emerge first. Let them grow until the seedlings of pot 2 emerge and produce a leaf.
7. Knock the soil out of the pots into a bucket of water and remove the seedlings from the soil, holding the soil and seedling under the surface of the water until the roots are free of soil.

Observations:

Compare and measure the length of the first internode (the distance from the corn seed to the beginning of the first leaf). Note that the internodes in both pots 1 and 2 stopped growing when the plant emerged from the soil; that is, when the seedling received light.

Supplementary Reading:

U.S. Agr. Res. Serv. New Light on Plants. U.S. Dept. Agr., Agr. Res. 1: 3-5. 1953.

DEMONSTRATION B-2: Control by light of growth and chlorophyll formation.

Materials:

1. A chamber or box that can be made completely dark. If entrance into the chamber cannot be made without exposing the contents to light, regardless of how weak the light is, then more than one chamber will

have to be made. These chambers can be made of masonite or plywood with caulked seams and a baffled door, or they can be made of several layers of black sateen cloth stretched over a wooden frame.

2. Two 40-watt fluorescent lamps.
3. Flats, boxes, pots, or plastic freezer cups filled with sterilized soil, sand, vermiculite, or perlite.
4. Bean seeds (any kind).

Procedure:

1. Plant the bean seeds and water. No nutrient solution is required even when the seeds are planted in sand, vermiculite, or perlite.
2. The best temperature is 80° to 85° F. Lower temperatures will suffice, but the rate of germination and growth will be slower.
3. Place the boxes in the dark chambers immediately after the beans are planted. If dark red kidney beans are used, planted in sand and kept at 80° to 85° F., they will germinate in 3 to 4 days.
4. On the fifth day, place box 1 in the light (preferably from fluorescent lamps) for 5 minutes, then return it to the dark chamber.
5. Repeat step 4 on the sixth, seventh, and eighth days from planting.
6. Place box 2 in the light for 20 minutes on the fifth day only.
7. Place box 3 under the light for 2 hours each day, and box 4 for a period of at least 8 hours per day.
8. Remove all boxes from the dark chambers on the ninth or tenth day from planting. Measure and record the length of each internode and the length of the leaves.
9. Slice or mince the leaves and place in a known volume of ethyl alcohol. Use the same volume of alcohol for each treatment irrespective of the size of the leaves. A better method is to use 10 milliliters of alcohol for each gram of leaves.
10. Calculate the average length of the internodes and the average length of the leaves for each treatment.
11. The relative amounts of chlorophyll can be estimated by assigning a numerical value to each sample based on the visual greenness of the extract, or the optical density of each sample can be measured.

Observations:

The plants grown in complete darkness (box 5) should have long hypocotyls, short first internodes, small leaves, and no chlorophyll. Boxes 1, 2, 3, and 4 should contain plants that have shorter hypocotyls, longer first internodes, and perhaps more internodes than the plants of box 5. They should also have much larger leaves. Plants of box 1 should contain no chlorophyll, and those of box 2 none or very little. Plants in boxes 3 and 4, however, should contain a greater amount of chlorophyll, with those plants of box 4 having more than those of box 3.

Supplementary Reading:

Downs, R. J. Photoreversibility of leaf and hypocotyl elongation of dark-grown red kidney bean seedlings. *Plant Physiol.* 30: 468-473. 1955.
Textbooks of Plant Physiology.

DEMONSTRATION B-3: Why plants bend toward light (phototropism).

Materials:

1. A chamber or box that can be made completely dark (see Demonstration B-2, steps 1 through 4 of Materials, for details).

Procedure:

1. Plant the bean seeds and water. No nutrient solution is required even when the seeds are planted in sand, vermiculite, or perlite.
2. The best temperature is 80° to 85° F. Lower temperatures will suffice, but the rate of germination and growth will be slower.
3. After the beans are planted, place one box in the dark chamber and one in the light, where the plants should receive 8 hours of light per day.
4. When the dark-grown beans are about 6 days old, open the door of the dark chamber so that the plants receive some light. Better results are obtained by placing a desk lamp 3 or 4 feet from the open door of the chamber. In a few hours these plants will bend toward the light entering the door.
5. When the plants in the light have expanded their first pair of leaves, place them in the dark chamber. This is conveniently done in the late afternoon. Again, open the door of the dark chamber and place a desk lamp 3 to 4 feet from the door.

Observations:

The following morning the leaf blades will have twisted around until they are perpendicular to the light.

Supplementary Reading:

Textbooks of Plant Physiology.

DEMONSTRATION B-4: Effect of red and far-red light on elongation of stems of light-grown plants.

Materials:

1. A light-equipped chamber (two 40-watt fluorescent lamps is a minimum).
2. A red chamber (a cardboard box with seams sealed with paper tape). Cut out the top and most of the bottom of the box. Place two layers of red cellophane over the opening in the bottom of the box, using cellophane tape to hold the cellophane in place.
3. A far-red chamber (a cardboard box prepared in the same manner as for the red chamber except cover the cut-out opening in the bottom with two layers of red and two layers of dark-blue cellophane).
4. A dark chamber.
5. Bean plants (preferably Pinto bean).

Procedure:

1. Plant beans in pots of sterilized soil, water, and place at a temperature of 80° F.
2. After 3 to 4 days the plants will begin to emerge from the soil. At this time all pots should be placed in the light chamber, where they should receive light from the fluorescent lamps for 8 to 10 hours each day. The temperature during this growing period should be 70° to 75° F.
3. The first pair of leaves should be about half expanded 10 to 12 days after planting. At this stage of development the plants are ready to start on treatments.
4. Divide the plants into three equal lots; A, B, and C.
5. Place plants of lot A in the dark at the close of the 8- to 10-hour light period. Place lots B and C under the red and blue cellophane (the far-red). Turn off the fluorescent lamps.
6. Place a 100-watt incandescent-filament lamp 3 feet from the red- and blue-cellophane filter and turn it on for 15 minutes.
7. Plants of lot B are moved in darkness to the dark chamber immediately after the 15-minute exposure to far-red. Plants of lot C are moved in darkness and placed under the box with the red-cellophane filter, which should be placed under the fluorescent lamps.
8. Turn on the fluorescent lamps for 10 minutes, then move plants of lot C in darkness to the dark chamber. (Great care should be taken to assure that the plants receive no light of any kind after they are exposed to red and to far-red.)
9. Return all plants to the fluorescent-light chamber the next morning.
10. The treatments should be given daily until a response is obvious, requiring at least 5 days of treatment.

Observations:

Record date of planting, date treatments were begun, number of days treatments were given, and the durations of the light period, red exposure, and far-red exposure. Measure and record daily the length of the second internode. Data can be plotted as line graphs (length plotted against time). Three plots should be made: The control (lot A), the far-red treatment (lot B), and the far-red followed by red (lot C).

Supplementary Reading:

Downs, R. J., S. B. Hendricks, and H. A. Borthwick. Photoreversible control of elongation of Pinto beans and other plants under normal conditions of growth. *Bot. Gaz.* 118: 99-208. 1957.
Wassink, E. C., and J. A. J. Stolwijk. Effects of light quality on growth. *Ann. Rev. Plant Physiol.* 7: 373-400. 1956.

C. Light and Plant Pigments

DEMONSTRATION C-1: Effect of light on formation of anthocyanin in seedlings.

Materials:

1. Seeds of turnip (Brassica rapa), variety Purple Top White Globe.
2. Five Petri dishes or plastic sandwich boxes with lids.

3. Filter paper (Whatman No. 3).
4. An aqueous solution of 32 parts per million chloramphenicol.
5. A solution of 0.01 molal HCl in aqueous 25-percent 1-propanol.
6. Black bags made of two layers of black sateen cloth.

Procedure:

1. Place three sheets of filter paper in each Petri dish and moisten with water and 32 parts per million chloramphenicol to depress chlorophyll formation.
2. Place 50 seeds of turnip on the filter paper, cover the dishes with the lids, and place each dish in a black cloth bag, keeping the temperature at about 77° F.
3. Allow 3 days for the seedlings to germinate and then begin treatments.
4. Place the dishes under the fluorescent lamps for periods of 0, 2, 4, 6, or 8 hours, then return the dishes to their black cloth bags.
5. The amount of anthocyanin formed can be observed after 16 to 24 hours after irradiation.
6. Count out a certain number of seedlings (say 25) from each dish and place each lot of 25 seedlings in the solution of 0.01 molal HCl in aqueous 25-percent 1-propanol. Place them in a refrigerator (about 40° F.) for 16 to 24 hours.

Observations:

The amount of anthocyanin formed by the various light treatments can be estimated by assigning a numerical scale that increases with increasing color, or it can be measured by differences in optical density as measured with a colorimeter or spectrophotometer. The amount of anthocyanin can then be plotted graphically by plotting the amount of anthocyanin as a function of the time (hours of light).

Supplementary Reading:

Siegelman, H. W., and S. B. Hendricks. Photocontrol of anthocyanin synthesis in turnip and red cabbage seedlings. *Plant Physiol.* 32: 393-398. 1957.

DEMONSTRATION C-2: Effect of light on tomato skin and fruit color.

Materials:

1. Mature green tomato fruits from a normally red-fruited variety when ripe, such as the variety Rutgers. Their skin color should be greenish-white with no visible red, pink, or yellow color. These are readily obtainable from home gardens any time prior to frost and in the larger cities southern-grown green-mature fruits can be obtained throughout the winter from local wholesale vegetable distributors.

2. A light facility, using incandescent or fluorescent lamps that provide a light intensity of 20 footcandles or more. Keep the temperature at 70° F.
3. A dark facility that provides total darkness, using light-tight black sateen cloth bags made of two layers of cloth. Keep the temperature at 70° F.
4. Scalpel or similar sharp instrument.
5. Small bottles or vials.
6. Acetone or petroleum ether (CAUTION: Flammable solvents.)

Procedure:

1. Divide the green-mature fruits into two uniform lots, A and B.
2. Ripen fruits of lot A for 10 to 14 days with illumination of daily duration of 1 hour. Longer periods may be given but are not necessary.
3. Ripen fruits of lot B 10 to 14 days in total darkness, taking care not to remove from darkness until ripe. These should ripen simultaneously with its lighted counterpart.
4. When the fruits are ripe, remove uniformly shaped and size sections from a typical fruit from each lot, being careful to keep the sections separate and properly identified as to treatment. Immerse each section in boiling water for 1 minute and cool immediately by immersion in cold water; thus the skin is readily removed from the tomato flesh. Scrape the adhering tissue from the skin with the scalpel as carefully and completely as possible. Place each scraped skin section in one of the small containers containing acetone or petroleum ether solvent and leach the skins with several washings (keeping the sections immersed) over a period of at least several hours.

Observations:

The fruits ripened in the dark will be pink; those ripened in the light will be orange-red. CAUTION: Fruits will ripen faster at temperatures higher than 70° F., but at those higher temperatures the red pigment in the flesh does not develop well and gives the fruit an off-color appearance.

The skin of fruits ripened in the dark will be colorless; the skin of those ripened in the light will have a yellow color even after prolonged leaching with the solvent. The insoluble light-induced yellow pigment left in the tomato fruit cuticle (skin) has not yet been identified. The presence or absence of the light-controlled pigment in the skins makes them either yellow or transparent. When the yellow skin is superimposed over the red, the fruit has an orange-red appearance, the typical coloration of summer field-ripened tomato fruits. The combination of red flesh and a transparent skin produces a fruit that is pink. The pink coloration is characteristic of fruits commercially available in the North in mid-winter which have been artificially ripened in darkness by vegetable wholesale distributors. If fruits ripened in the dark have yellow-tinted skins, the leaching process was not complete or the fruits were too mature and were already producing the light-responsive pigment at the time the fruits were placed in the dark. Light starts to act as soon as the fruits mature.

Supplementary Reading:

- Piringer, A. A., and P. H. Heinze. Effect of light on the formation of pigment in the tomato fruit cuticle. *Plant Physiol.* 29: 467-472. 1954.
U.S. Agr. Res. Serv. Light Link in Tomato. U.S. Dept. Agr., Agr. Res. 2: 8. 1954.

DEMONSTRATION C-3: Localization of response to light by the pigment in tomato skin.

Materials:

1. Use the same materials as in Demonstration C-2.
2. In addition, two small sheets of aluminum foil, enough to completely cover a tomato fruit.

Procedure:

1. Carefully select three uniform green-mature fruits.
2. Completely and tightly wrap one of the tomato fruits with a sheet of aluminum foil (dark control).
3. Remove a 5-millimeter-diameter section from the center of another sheet of aluminum foil. Completely and tightly wrap another tomato fruit, being careful that the perforation exposes an area of skin on the side of the fruit.
4. Leave the remaining tomato fruit unwrapped (light control).
5. Place all tomato fruits, both wrapped and unwrapped, in the light and allow 10 to 14 days for ripening, keeping the temperature at 70° F.
6. When the unwrapped fruit is ripe (soft and red), unwrap all fruits and note any differences in skin color.
7. Sections of the skin should be removed and processed as in Demonstration C-2, being careful to include the area exposed to the light through the perforation in the foil.

Observations:

Skins of the unwrapped fruit should be bright yellow; skins of the completely wrapped fruits should be colorless; skins from the wrapped fruit with the small area exposed should be colorless except for the small exposed area which will be yellow. A novelty can be produced by tightly wrapping a green-mature fruit with a sheet of aluminum foil having a number of small perforations.

Supplementary Reading:

- Hicks, C. B. You can make a plant do tricks. *Popular Mechanics* 108: 81-85, 232-236. 1957.

DEMONSTRATION C-4: Effect of light on coloration of apples.

Materials:

1. Early harvested (green) Jonathan, Rome Beauty, or Arkansas apples. Jonathan variety is preferred. Store the apples at 32° F. in bags of 0.38-millimeter polyethylene plastic.
2. Black cloth bags.
3. Black plastic electrical tape.
4. Aluminum foil.

Procedure:

1. Place one apple in a black cloth bag.
2. Wrap one apple in aluminum foil, then cut holes in the foil and place the apple under the light of the growth chamber.
3. Using the black plastic electrical tape, put an initial on each of several apples and place the apples under the light of the growth chamber.
4. Allow 3 to 4 days for the apples to turn red, then remove the foil, tape, or black bag. Keep the temperature about 75° F.

Observations:

The apple that has been kept in the dark will still be green. The one covered with aluminum foil will also be green except where light has entered through the cut-out holes; here, the apple will be red and may have a polka-dot appearance of red on green. The apple that was exposed to light except for areas under the black tape will be red. Under the black tape the apple will be green and thus show green initials on a red apple.

Supplementary Reading:

U.S. Agr. Res. Serv. How Light Controls Plant Development. U.S. Dept. Agr., Agr. Res. 8: 3-5, 1959.

Siegelman, H. W., and S. B. Hendricks. Photocontrol of anthocyanin synthesis in apple skin. Plant Physiology 33: 185-190. 1958.

D. Duration of Light

DEMONSTRATION D-1: Photoperiodic control of flowering of short-day plants.

Materials:

1. A light-equipped chamber (two 40-watt fluorescent lamps is a minimum).
2. A dark chamber.

3. Plants of cocklebur, lambsquarter, scarlet sage variety America, or morning-glory variety Scarlett O'Hara, should be grown on daylengths of 18 hours or more until large enough to use in the demonstration. Use morning-glory plants as soon as the cotyledons have expanded. Plants of cocklebur and lambsquarter are large enough when they have three leaves above the cotyledons. Photoperiodic treatments of scarlet sage can be begun as soon as the plants have 4 to 5 pairs of leaves.
4. Sterilized soil.

Procedure:

1. When the plants are large enough to use, divide them into lots A and B.
2. Both lots of plants should receive 8 to 10 hours of light in the light chamber each day.
3. Place lot A in darkness at the close of each daily light period. Turn off fluorescent lamps. Place lot B 3 to 4 feet from a 40-watt incandescent-filament lamp, which is now turned on.
4. If an electric time switch is available, give lot B a total light period of 18 to 20 hours (8 to 10 hours fluorescent light and 8 to 10 additional hours incandescent light).
5. If an electric time switch is not available, leave the incandescent lamp on throughout the night.
6. Return both lots A and B to the light chamber each morning.
7. Continue these daily treatments until flower buds are obvious. If the treatments are discontinued now, the flowers will usually continue to develop. During this period of development both lots of plants should receive photoperiods of 18 hours or more.

Observations:

Record date of planting, date treatments began, length of the light and dark periods, and include the number of short days required to induce flower formation.

Supplementary Reading:

- Doorenbos, J., and S. J. Wellensiek. Photoperiodic control of floral induction. *Ann. Rev. Plant Physiol.* 10: 147-184. 1959.
- Lang, A. Physiology of flowering. *Ann. Rev. Plant Physiol.* 3: 265-306. 1952.
- Liverman, J. L. The physiology of flowering. *Ann. Rev. Plant Physiol.* 6: 177-210. 1955.
- Naylor, A. W. The control of flowering. *Scientific American* 186: 49-56. 1952.
- Parker, M. W., and H. A. Borthwick. Influence of light on plant growth. *Ann. Rev. Plant Physiol.* 1: 43-58. 1950.
- Salisbury, F. B. The flowering process. *Scientific American* 198: 109-117. 1958.

DEMONSTRATION D-2: Photoperiodic control of flowering of long-day plants.

Materials:

1. A light-equipped chamber (a minimum of two 40-watt fluorescent lamps).
2. A dark chamber.
3. Plants of tuberous-rooted begonia, petunia, or barley.

Procedure:

1. Divide the plants into lots A and B as soon as they emerge from the soil.
2. Both lots should receive 8 to 10 hours of light daily in the light chamber.
3. Place lot A in darkness at the close of each 8- to 10-hour light period. Turn off fluorescent lamps and expose plants of lot B to light from a 40-watt incandescent-filament lamp.
4. If an electric time switch is available, allow a total light period of 16 to 18 hours, 8 to 10 hours from the fluorescent lamps and another 8 hours from the incandescent lamp.
5. If an electric time switch is not available, leave the incandescent lamp on throughout the night.
6. Return both lots to the fluorescent-lighted chamber each morning.
7. These treatments should be given daily until flower buds are obvious. If the treatments are stopped at this point, the flower buds will usually continue to develop. During this period of development all plants should be grown on short days of 8 to 10 hours' duration.

Observations:

Record date of planting, date demonstration began, length of the light and dark periods, and also record how many long days were required to induce formation of flowers. If the tuberous-rooted begonia is used as the experimental plants, make observations on the extent of tuber formation as well as flowering.

Supplementary Reading:

U.S. Agr. Res. Serv. Prescription for Better Plant Form. Dept. of Agr., Agr. Res. 8: 14. 1959.
See Demonstration D-1.

DEMONSTRATION D-3: Photoperiodic control of growth and dormancy of woody plants.

Materials:

1. A light-equipped chamber.
2. A dark chamber.
3. Seedlings or rooted cuttings of some woody plant material such as deciduous trees (catalpa and red maple), evergreen trees (spruce, loblolly pine, slash pine, or Virginia pine), and shrubs (hollies and Weigela).

Procedure:

1. Divide rooted cuttings or seedlings into lots A and B and place both lots in the light chamber for 8 to 10 hours daily.
2. Place lot A in darkness at the close of each 8- to 10-hour light period.
3. Turn off the fluorescent lamps and place lot B 3 to 4 feet from a 40-watt incandescent-filament lamp, which is now turned on.
4. If an electric time switch is available, the total daily light period for plants of lot B should be 16 to 18 hours, 8 to 10 hours of fluorescent light plus 8 more hours of incandescent light.
5. If an electric time switch is not available, leave the incandescent light on throughout the night.
6. Return both lots A and B to the light chamber each morning.
7. These daily treatments should be continued until the plants on 8- to 10-hour day (those of lot A) take on the aspects of dormancy and there is a marked difference in the size of plants of lots A and B. This should require at least 30 days.

Observations:

Record date treatments began, length of the light and dark periods, and also record the number of short days required to induce dormancy or to stop growth of the main axis. Measurements can be made at daily intervals and the length of the main axis of plants from both lots can be plotted against time in days.

Supplementary Reading:

- Borthwick, H. A. Light effects on tree growth and seed germination. *Ohio Jour. Sci.* 57: 357-364. 1957.
- Downs, R. J., and H. A. Borthwick. Effects of photoperiod on growth of trees. *Bot. Gaz.* 117: 310-326. 1956.
- Thimann, K. V. The physiology of forest trees. Ronald Press, New York, N. Y., pp. 529-583. 1958.
- Wareing, P. F. Photoperiodism in woody plants. *Ann. Rev. Plant Physiol.* 7: 191-214. 1956.

DEMONSTRATION D-4: Photoperiodic control of bulb formation of onions.

Materials:

1. Onion seeds. Plants of southern varieties White Bermuda, Crystal Wax, Eclipse, Excel, and Granex hybrid will bulb on 12-hour days. Plants of northern varieties Australian Brown, Sweet Spanish, Elite hybrid, and Yellow Globe (Early Yellow Globe, Yellow Globe Danvers, Downing Yellow, and Globe) will bulb on 15-hour days.
2. Sterilized soil.

3. Four wooden boxes 8 to 10 inches wide, 10 inches deep, and 12 inches long with drainage holes.
4. A light-equipped chamber.
5. A dark chamber.

Procedure:

1. Fill boxes with sterilized soil and level the soil surface.
2. Make two shallow furrows lengthwise of the box 1/4-inch deep and 4 inches apart.
3. Plant the seeds thinly in the furrows, cover the seeds with soil, and water carefully. Label each box with the name of the variety, the date of planting, and the daylength treatment.
4. When seedlings are well established, thin the plants to 2 inches apart in the row.
5. Germinate the seeds and grow the plants at room temperature (70° to 80° F).
6. Place two boxes on short days and the remaining two boxes on long days immediately after planting the seeds.
7. Additional varieties and intermediate daylengths can be used to broaden the experiment.

Observations:

Note any differences in the top growth or plant habit at regular intervals during the course of the demonstration. Differences should become apparent in about 60 days. Carefully remove a few plants at random from a box on each daylength and note any differences in bulbing. Do this at regular intervals to determine the time of bulbing and the treatment on which it occurred. When bulbing is definitely apparent, the experiment may be terminated. The plants in the remaining box on each daylength may be harvested and the extent of bulbing on each daylength noted and recorded.

Supplementary Reading:

- Boswell, V. R., and H. A. Jones. Climate and vegetable crops: Onions. In CLIMATE AND MAN, U.S. Dept. of Agr., Yearbook of Agriculture, 1941. Government Printing Office, Washington, D.C., pp. 388-389.
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