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Light and Plant Development

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INTRODUCTION

Each plant is the product of its genetics and the total environment in which it is grown. Some grow in harsh environments whereas others grow under milder conditions. Some plants complete their life cycle from seed germination through production of the next generation of seed within a single year (annuals). Others germinate and begin growth one year, survive over winter, and produce seed during the second year (biennials). Still others grow for many years and produce many crops of seed (perennials). Regardless of its growth habitat, the "strategy" of each plant is to survive in its environment long enough to produce the next generation. Therefore, the plant must be able to sense various aspects of its total environment and activate or repress genes that regulate adaptation of the plant to the environment in which it is growing.

The total growth environment is constantly changing during the lifetime of a plant. It includes available soil moisture, quantities and solubilities of mineral nutrients, acidity of the soil solution, diseases, insects, air and soil temperatures, and light. Because of the growth and developmental processes associated with adaptation to the many environmental variables, plants of the same genotype can differ significantly in size and chemical composition.

The most widely studied aspect of light–plant relationships is photosynthesis, in which the energy of light is absorbed by the growing plant

and used in combining carbon, hydrogen, and oxygen into simple sugars. However, an equally important aspect of plant survival and productivity is the partitioning and use of that photosynthate within the plant. This chapter will concentrate on aspects of plant growth and development called photomorphogenesis, as influenced by photoperiod (day length), light quantity, and light quality. Some of the light-regulated processes are modified by temperature. Many of the examples used in this chapter are from research conducted by the author and his colleagues from the late 1950s to the present time.

PHOTOMORPHOGENESIS

In addition to its very significant role in photosynthesis, light is involved in natural regulation of how and where photosynthate is used within a developing plant. Plants contain photoreceptor systems that sense, or measure, various aspects of the light environment and initiate physiological processes that regulate adaptation of the plant to increase its probability of survival and reproduction in that environment. Plants respond to day length as an initiator of seasonal events, such as flowering or development of fleshy roots and tubers. They also respond to light quality (spectral distribution) and light quantity (photon flux density). Under experimental conditions in controlled environment chambers, each of these factors can be studied in detail while the other environmental components are held constant. However, in the real world plants grow in constantly changing total environments, including natural day lengths and changes in light quantities associated with season, time of day, and competition from other plants. Light spectral distribution is also influenced by reflection from competing plants. Even reflection from different colored soils, plant residues, and mulches can affect morphological development. This chapter will concentrate on five main phases in the accumulation of knowledge on light regulation of plant development: (a) discovery of photoperiodism and its significance, (b) discovery of a photoreversible pigment system, phytochrome, (c) phytochrome-regulated developmental responses under controlled environments, (d) the importance of far-red light reflected from other plants under field conditions, and (e) the theory and use of colored soil covers (mulches) to regulate spectrum of upwardly reflected light and its affect on field-grown plants.

Photoperiodism

The research that led to the discovery of photoperiodism was begun in the early 1900s by W. W. Garner and H. A. Allard, who worked with

the Maryland type of tobacco (*Nicotiana tabacum* L.). They conducted research on the old U.S. Department of Agriculture farm at Arlington, Virginia, close to where the Pentagon now stands. Research was less specialized at that time and the same scientist often studied a broad range of plant problems. Thus, the research of Garner and Allard involved various aspects of crop production including plant nutrition, virus and disease resistance, and cultivation of tobacco and some other agricultural plants. The program also involved cross-breeding and development of improved genetic lines and varieties.

Genetic materials with desired disease resistances and other characteristics were grown in field plots and evaluated for possible use in cross-breeding combinations. The tobacco being evaluated in the field included some "mammoth" plants that developed many leaves but flowered long after the other genetic lines had set seed. According to Garner and Allard (1), some mammoth (initially called "giant") plants were observed in the field as early as 1906. Since leaf production is important in tobacco, they became interested in possible incorporation of the leaf factor into some of their genetic combinations. The fact that flowering of the mammoth strain was not synchronized with flowering of other desirable strains was a problem. As was common practice among tobacco breeders, some potentially useful plants were transferred from the field to a greenhouse in autumn before the first killing frost. The intact mammoth plants flowered and set seed in the greenhouse, as did the regrowth from stumps of plants that were cut back before transfer to the greenhouse. The procedure was cumbersome, but it was possible to produce seed. One of the existing concepts was that the mammoth strain had to be older than other strains before it could flower. One year, some seeds of the mammoth strain were sown very early in the greenhouse so that the plants would be old enough to flower in the field at the same time as the other genetic lines. Contrary to the plant age hypothesis, the early-sown plants flowered in the greenhouse at a relatively small size. Clearly, something about the greenhouse conditions in winter resulted in altered time of flowering. Garner and Allard suspected that the number of hours of light per day had something to do with timing of flower development. Another possibility was lower temperature. They tested the day length concept by giving some plants extra hours of light each day. Other treatments consisted of moving some plants into darkness before sunset to shorten their day length. The scientists suspected that other plant species might also respond to day length, and the studies included soybean [*Glycine max* (L.) Merr.] and some other species in addition to the "Maryland Mammoth" strain of tobacco. The classic paper that described the discovery of photoperiodism was published by Garner and Allard in 1920 (1).

Some of the "tools" used in Garner and Allard's research are shown in Figure 1. The upper photograph shows a box of field-grown soybean plants being moved into a dark chamber during the day. In this way it was possible to determine whether a shorter-than-natural day length would alter time of flowering. As a historic note, the first treatment in the dark chamber began at 4 p.m. on July 10, 1918, when a box of the "Peking" cultivar of soybean and three pots of Maryland Mammoth tobacco were placed in the ventilated dark chamber (1). The plants were removed from the dark chamber at 9 a.m. the next morning and the sequence was repeated each day until the seeds of soybean and tobacco were mature. Plants that received the shortened day treatment in the dark chamber matured earlier than control plants left on natural days. Treated plants were moved into and out of the dark chamber by hand during that initial experiment in 1918.

A larger "dark house" was constructed the following spring (Fig. 1, lower photograph). It was designed for easier moving of plants into and out of the dark chambers (1). There were four steel tracks, each entering the building by a separate door. Low-platform trucks were mounted on the tracks to allow the boxes of plants to be moved into or out of the dark chambers. By utilizing different chambers within the building, it was possible to give several different light/dark (day length) combinations at the same time. For example, some treatments involved darkness from 4 p.m. to 9 a.m. whereas others were in darkness from 6 p.m. until 6 a.m., etc. Some were even moved into darkness at 10 a.m. and back to daylight at 2 p.m. to break the natural day into two shorter days.

Other experiments during the autumn of 1919 utilized greenhouses to compare flowering of Maryland Mammoth tobacco on natural winter day lengths with similar plants grown on natural winter day lengths that were extended from 4:30 p.m. until 12:30 a.m. with supplemental light from tungsten filament lamps.

The combination of experiments with tobacco and soybean compared plant responses to shortened vs. natural day lengths in summer and natural vs. extended day lengths in winter. Maryland Mammoth plants flowered earlier and at smaller size when grown on short days. Garner and Allard (1) suggested the term *photoperiod* to describe length of day and *photoperiodism* to describe the response of an organism to the relative length of day. Many subsequent experiments were done with many plant species.

Extending the natural day length with supplemental light resulted in delayed flowering in species such as tobacco, soybean, and cocklebur (*Xanthium pensylvanicum* Wallr.). The term "short-day plants" was coined to describe this group because their flowering time was hastened

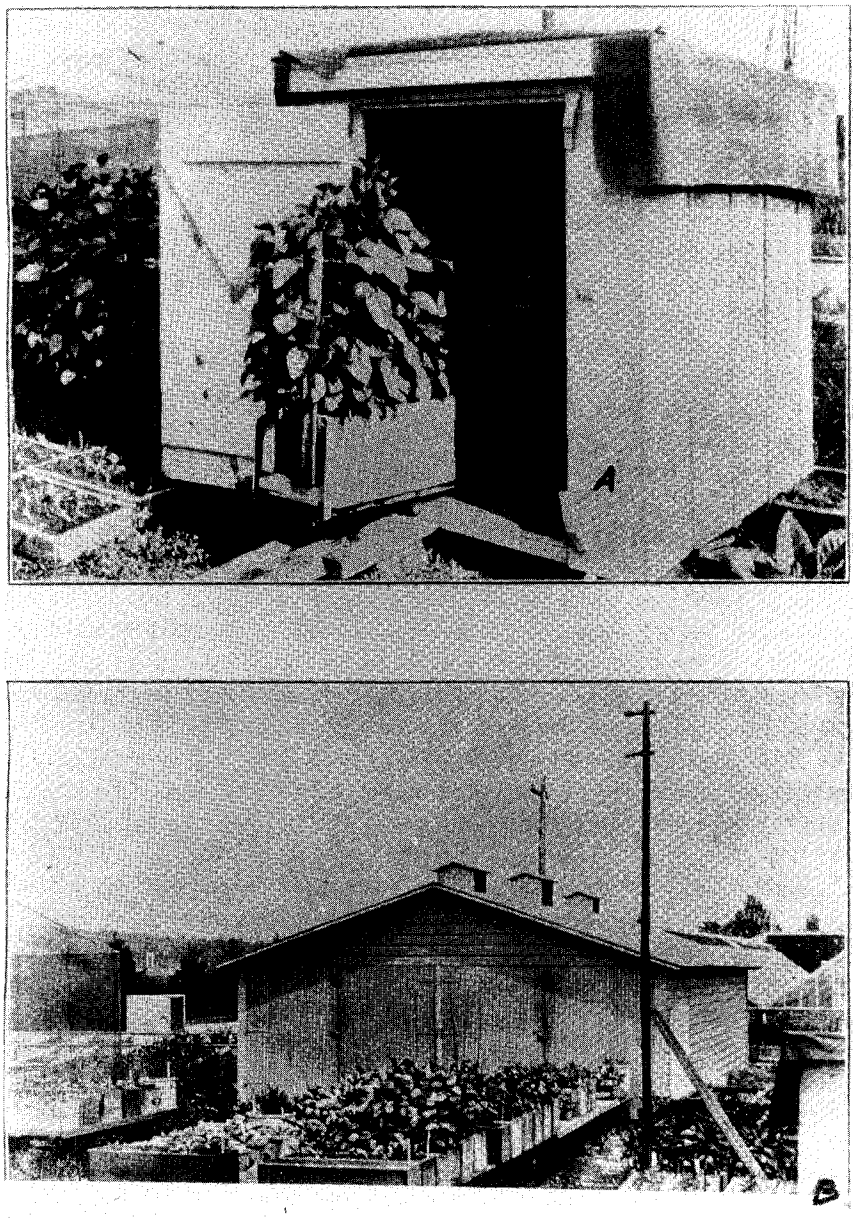

Figure 1 "Tools" used by W. W. Garner and H. A. Allard in the discovery of photoperiodism: dark chamber used in 1918 (upper photograph) and the larger "dark house" used in 1919 (lower photograph).

by short days and delayed by long days. Time of flowering of some other plants, such as barley (*Hordeum vulgare* L.), was hastened if the plants received supplemental light to extend the natural days. This group was called "long-day plants." A third category appeared to be indifferent to day length and became known as "day-neutral" types.

The discovery of photoperiodism was highly significant for plant breeders who could then use supplemental light to hasten and synchronize flowering time of long-day plants. They could also use supplemental lighting to keep short-day plants vegetative for a while, then provide short days by moving them into dark rooms (or covering them with light-tight curtains) from about 5 p.m. until 8 or 9 a.m. to synchronize flowering time for short-day species.

A dramatic example of photoperiodic control of flowering of Maryland Mammoth tobacco is shown in Figure 2. Both of the plants grew from the same lot of seed supplied to this author by Dr. James E. McMurtrey, Jr. in 1962 when he was in charge of the USDA tobacco physiology research (about 40 years earlier he was a junior colleague of Garner and Allard). The small plant was grown on 8-hr days alternated with uninterrupted 16-hr nights. When photographed, it was about 0.6 meters tall, had 23 leaf nodes, and had already flowered and set seed. The large plant was grown in a greenhouse that received natural day lengths plus several hours of supplemental light so that it always received long days. Even though the large plant was started earlier than the small one, it had grown to a height of more than 4.5 m, had more than 190 leaves, and had not yet flowered when this photograph was taken. Shortly thereafter, the plant grew beyond the supplemental light fixture and flowered.

Following the classic discovery by Garner and Allard, many scientists throughout the world published papers showing that other species sensed photoperiod and used that environmental signal to initiate flowering. As the papers appeared, it became apparent that the photoperiod-sensing mechanism was sometimes highly modified by temperature. Also, after the term photoperiod (for day length) was firmly established in the scientific literature, it became apparent that the number of hours of uninterrupted darkness rather than the hours of light was the dominant factor involved in the timing mechanism (2). The next major step in the research was based on the fact that a short period of darkness during the day did not affect flowering time, whereas a short period of light near the middle of the night delayed flowering of short-day plants and hastened flowering of long-day plants.

Figure 2 Maryland Mammoth plants grown on 8-hr days (small plant with seed) or in a greenhouse with natural day lengths plus several hours of supplemental light to provide long-day treatment (tall plant).

Discovery of Phytochrome

In the mid-1930s, a new USDA research team was organized at Beltsville, Maryland, to study the nature of photoperiodism and its significance to agriculture. The new team was headed by Harry A. Borthwick (a botanist) and Marion W. Parker (a plant physiologist). The approach was to discover the light-sensing mechanism involved in photoperiodic control of flowering and other aspects of plant development. Two "photoperiod houses" were constructed at Beltsville. They were similar to the earlier "dark house" at Arlington in that boxes of plants were mounted on carts and moved into and out of the buildings on steel rails. The new buildings were equipped with electricity, and light-tight curtains were used to separate treatment compartments within the buildings. This allowed use of natural outdoor daylight alternated with various timing and light combinations when the plants were inside the photoperiod houses.

Some of the research was done in greenhouses equipped with various supplemental light sources and adjacent dark rooms. The plants were grown on warehouse carts rather than on fixed benches in order to allow more orderly movement to adjacent rooms for various supplemental light and temperature combinations during the night.

Other aspects of the research required a more completely controlled light environment and resulted in construction of artificially illuminated growth rooms so that light intensity during the daily light period would not vary with season, as it did in the greenhouse and outdoors next to the photoperiod houses. In order to obtain adequate light for plant growth in those early rooms, the team used a carbon arc lighting system supplemented with white incandescent filament lamps arranged in a circle around the carbon arc (Fig. 3). The table used to support growing plants was also circular in shape and placed below the incandescent lamps (see Fig. 3). The carbon arc system was surrounded by glass in order to filter out the ultraviolet light before it reached the plants. Occasionally a window broke and an experiment was ruined. However, this lighting system was the best available at the time of its construction in 1937 (3), and it was used successfully until 1962 (4), when it was replaced by a combination of white fluorescent and incandescent filament lamps. The carbon arc growth room was instrumental in development of the 8-hr light period as the standard "short day." This came about quite naturally because the carbons would burn for about 8 hr and 15 min before needing replacement. Thus, many of the early growth room experiments with soybean and cocklebur (both short-day plants) involved 8 hr of the bright light, and other light combinations given in adjacent rooms where the

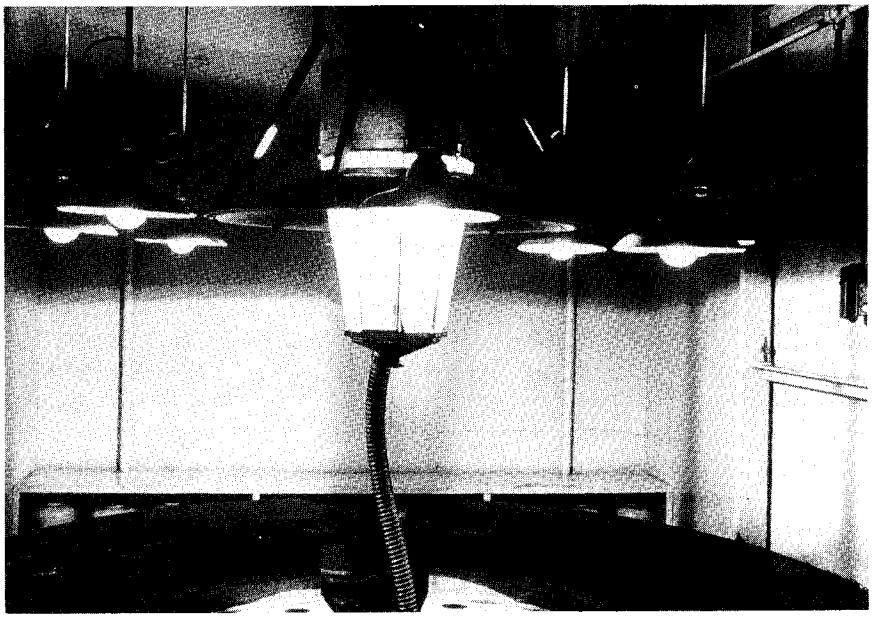

Figure 3 An early plant growth room at Beltsville illuminated with a carbon arc lamp supplemented with incandescent filament lamps.

plants were treated with various colors, durations, and intensities of light during the 16-hr night.

While some experiments were done in the artificially illuminated growth rooms, others were done with a combination of outdoor daylight and timing of supplemental light during the night in the photoperiod houses. Still other experiments were done in the greenhouse. Some of these experiments were designed to test which color of light was most effective as a night interruption. The rationale was that the effectiveness of different colors would indicate absorption characteristics of the pigment system involved in the photoperiodic responses of plants, and responsiveness to timing of the supplemental light would provide evidence toward a mechanism of action.

Also in the 1930s, scientists at the Smithsonian Institution in Washington, DC, constructed a number of small irradiation chambers that allowed testing the effects of narrow wavebands of light on seed germination. At that time, the Smithsonian research group worked within the "castle" building on the Mall and the USDA seed research was conducted

about a block away in the USDA building. Flint, of the USDA, and McAllister, of the Smithsonian Institution, experimented with a selection of lettuce (*Lactuca sativa*) seed that had a low percentage of germination in uninterrupted darkness but a high percentage if exposed to white light (5,6). Using a series of fixed filter chambers, they found that germination of this selection of seed was increased by exposure to wavelengths at about 660 nm. However, germination was decreased below that of the dark controls after exposure to wavelengths at about 730 nm.

At Beltsville, Borthwick, Parker, and colleagues experimented with whole intact plants using white light filtered through broad-band colored glass filters at various times during the night. They found that the red component of white light was most effective in regulating time of flowering. After confirmation of the effectiveness of red light with a number of plant species, Borthwick and Parker decided to conduct quantitative studies on involvement of light color in regulation of the flowering process. It was decided that the approach would require action spectra (i.e., efficiency of different wavelengths of light) for control of flowering as a means of learning more about the light sensor within the plant. Sterling B. Hendricks (a physical chemist interested in botany) joined the group and built a double-prism spectrograph largely from surplus items and two prisms borrowed from the Smithsonian Institution. The theory, design, and construction of the spectrograph were evidence of the combined innovation, resourcefulness, and scientific genius of Sterling Hendricks, Harry Borthwick, and Marion Parker. The light source was a 12-kW carbon arc projector that was once used to provide the "spotlight" on the stage of a nearby theater. (Hendricks explained to this author that it was Parker who "rescued" the carbon arc projector as it was being discarded by a burlesque theater in Baltimore.) The prisms borrowed by Hendricks from the Smithsonian Institution were also historic. They had been used by Samuel Pierpont Langley (1834–1906), a noted astronomer, physicist, and aeronautics pioneer. A diagram of the "Beltsville spectrograph" is shown in Figure 4.

During operation of the spectrograph, the beam of light from the carbon arc first passed through a narrow vertical slit to a front-surfaced mirror, then through the prisms and through a door (which served as a manually operated shutter) to the treatment table. Large plants such as soybean and cocklebur were trimmed to a single leaf and that leaf was placed in a specific waveband, as diagrammed in Figure 4. The procedure for small plants such as *Chenopodium* required a modification. For these materials, the "rainbow" of colors was beamed at a front-surfaced mirror above the treatment table and reflected downward onto the seedlings. The treatment table was movable and could be placed

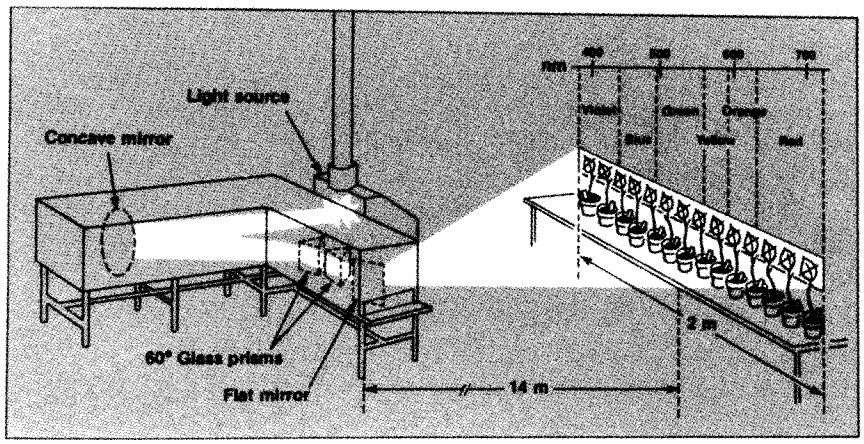

Figure 4 Diagram of the Beltsville spectrograph showing path of light from carbon arc through prisms to treatment table where soybean plants trimmed to a single leaf are being treated.

closer to or farther from the spectrograph. Increasing the distance allowed greater resolution of wavebands but decreased photon flux density at a given point on the treatment table. The spectrograph was used in development of many action spectra for control of flowering, seed germination, leaf movement, stem elongation, and many other morphological responses by scientists from around the world. It was a very important "research tool" from the time of its construction in the 1940s (7) until it was dismantled by Hendricks just before he retired in 1970.

The spectrograph separated white light into its component colors and allowed exposure of plants to a range of wavelengths for various durations, usually near the middle of the night following 8 hr of bright light in the nearby growth room. To obtain an action spectrum for control of flowering in soybean, for example, it was necessary to first determine how old the plants had to be before they were florally responsive to photoperiod, and how many short days were needed to cause flowering of "control" plants that received 16-hr uninterrupted nights alternated with 8-hr light periods (7). It was also necessary to determine whether plants such as soybean or cocklebur could be trimmed to a single leaf so that the treated part of the plant could be put in exactly the same position in the spectrum each day for the three or four consecutive days of treatment (see Fig. 4). In order to get different energy levels at each waveband, it was necessary to use different exposure durations. For example, one set of

plants would be arranged as shown in Figure 4, irradiated in the "rainbow" of colors for one minute and then returned to darkness. Then another set would be put in the same positions for 2 min, followed by return to darkness. Other sets of identically pretreated plants received 4-, or 8-, or 16-, or 32-min treatments before returning to darkness. The same procedure with the same plants had to be done for several consecutive days. The early action spectra for control of flowering (7,8) were based on relatively few plants, and the red action peak seemed to be a broad band from about 640 to 660 nm. Germination of light-requiring seed offered the possibility of greater precision because the small size of seeds (relative to the size of intact plants) allowed use of more experimental units (seeds) on the spectrograph.

Borthwick et al. (9) tested germination of lettuce seeds and found a prominent peak for promotion of germination near 660 nm and a depression of germination near 730 nm. When seeds that had been pretreated with enough red light to promote germination were placed on the spectrograph, the scientists found an inhibition peak near 730 nm. They hypothesized that the effect of red light at 660 nm was reversed or negated by exposure to far-red (then called near-infrared) at 730 nm. To test the theory, they irradiated seeds repeatedly with red and far-red (Table 1). It was apparent that the effects of red could be reversed by far-red and vice versa. This observation became a very important step toward discovery of a photoreversible regulatory system. In subsequent spectrographic experiments with whole plants, the plants were often irradiated for a few minutes with red light before placement in the far-red

Table 1 Germination Responses of Grand Rapids Lettuce Seeds to Repeated 1-min Irradiations with Red (R) Alternated with 4-min Irradiations with Far-red (FR) Light

Irradiation	Germination (%)
None (dark control)	9
R	98
R, FR	54
R, FR, R	100
R, FR, R, FR	43
R, FR, R, FR, R	99
R, FR, R, FR, R, FR	54
R, FR, R, FR, R, FR, R	98

Source: Adapted from Ref. 9.

part of the spectrum in order to test action spectra for photoreversible control. The far-red action peak was near 730 nm for control of seed germination and floral induction in whole plants. However, an apparent discrepancy between red action peaks for seed germination and control of flowering was noted. The red action peak in whole plants appeared to be near 650 nm (7,8), whereas the peak for seed germination was near 660 nm (9).

The research on photoreversible control of seed germination (9), stem elongation (10,11) and hypocotyl hook opening (12) provided evidence of the presence of a photoreversible pigment system that responded to low energies of red and far-red light. It was apparent that one form of the pigment absorbed red light and became the far-red–absorbing form, which then absorbed far-red and became the red-absorbing form, etc.

With information drawn from the spectrographic studies, reversible responses to red and far-red, and the expertise of K. H. Norris (an engineer), W. L. Butler (a physicist), and H. W. Siegelman (a chemist), the team was able to build a dual-wavelength photometer and to photometrically determine the presence of a red/far-red photoreversible pigment in dark-grown seedlings. The paper was published by Butler et al. (13) in the *Proceedings of the National Academy of Science USA* in 1959. As other studies were published, the name *phytochrome* became firmly established.

After the discovery of phytochrome, there was an explosion of interest in laboratories around the world. Many followed the lead of Siegelman and Hendricks and concentrated on purification and identification of phytochrome from dark-grown etiolated seedlings. Preliminary studies showed that dark-grown seedlings contained more phytochrome and obviously less chlorophyll (which also absorbs red light) than light-grown seedlings. Some scientists questioned whether the "first phytochrome" in dark-grown seedlings would be the same as that in light-grown green plants. However, extraction from dark-grown seedlings offered more promise of success than extraction from green plants. The rationale was that if one knew the chemistry of phytochrome it would lead to better understanding of regulatory mechanisms within growing plants. Although many studies have been conducted, progress toward chemical characterization of phytochrome was much slower than originally envisioned by Hendricks.

Other scientists, including this author, concentrated on phytochrome regulation of various processes and endproducts in green plants. The objectives of that approach were to learn about the basic regulatory action of phytochrome in growing plants and to use that information to improve quantity and quality of plant products in a crop production sys-

tem. The approach was to begin with precise spectrographic studies, follow these with observations of real crop production problems in the field, then use controlled environments and broad-band red and far-red light sources to study plant responses, and to relate these phytochrome-mediated responses to field growth under modified production practices. This author joined the Beltsville group in 1961 on a postdoctoral fellowship after completing doctoral research with Walter E. Loomis at Iowa State University on interactions of photoperiod and temperature on flowering and shoot/root partitioning in a biennial legume plant, sweetclover (*Melilotus alba* L.). It was apparent during the Iowa research that some photoperiodic responses such as flowering differed with temperature, and others such as shoot/root biomass ratios in first-year biennial plants were dominated by photoperiod (Table 2). After arriving at Beltsville, the first step was to adapt and use small-size seedlings to develop highly refined action spectra on the spectrograph. An Iowa strain of pigweed (*Amaranthus retroflexus* L.) and a Canadian strain of *Chenopodium rubrum* (L.) were compared for early responsiveness. Both could be induced to flower on short days (alternated with uninterrupted long nights) soon after emergence. However, the *Chenopodium* was responsive in the cotyledonary stage (Fig. 5). Thus, the series of experiments was done with this species. More than 100 *Chenopodium* seedlings could be placed in the same space needed for one soybean or cocklebur plant. Use of the miniplant system greatly improved precision of the action spectra and quickly became a team effort with Borthwick and Hendricks. A tray of *Chenopodium rubrum* seedlings old enough for treatment on the spectrograph and a seedling of the Beltsville strain of cocklebur that is also old enough for treatment are shown in Figure 6. For treatment, the *Chenopodium* seedlings were thinned to 10 per row and the rows were

Table 2 Shoot/Root Biomass Ratios in First-Year Biennial Sweetclover Plants Grown Under Three Photoperiods from July 15 until November 15 in a Greenhouse and Outdoors[a] at Ames, Iowa (42°N Latitude)

Photoperiods and locations				
24 hr			9 hr	
Greenhouse	Outdoors	Natural Outdoors	Greenhouse	Outdoors
(shoot/root biomass ratios)				
3.9	3.8	0.5	0.4	0.3

[a] Outdoor temperatures were lower than those in the greenhouse late in the season.
Source: Adapted from Ref. 14.

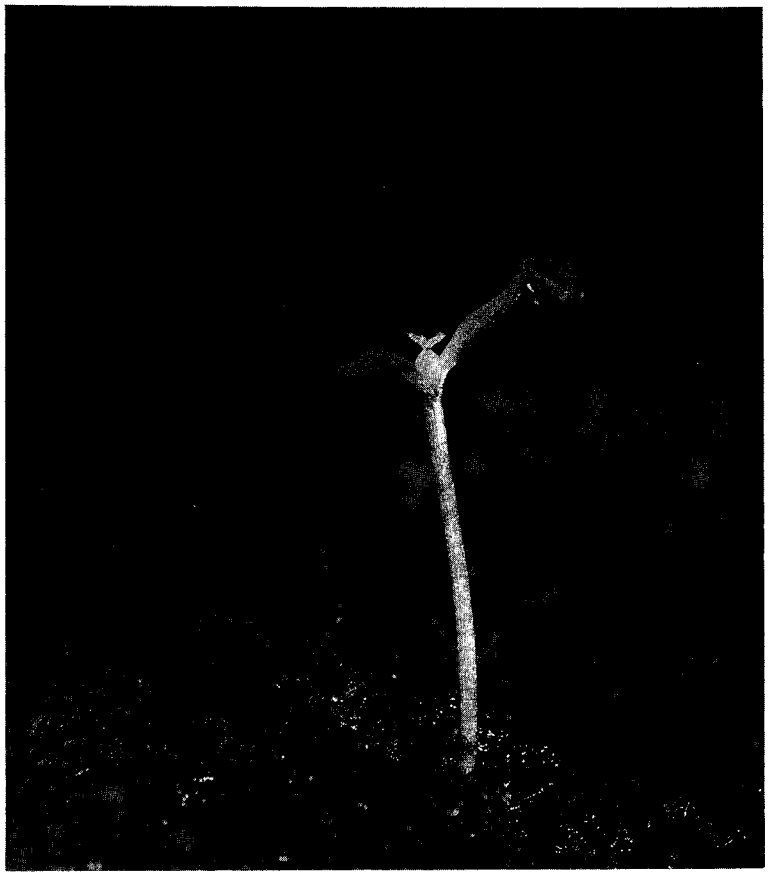

Figure 5 A *Chenopodium rubrum* seedling that was induced to develop a single floret between its cotyledons, without leaf formation. Note that the seed coat is still evident.

about 1.5 cm apart. A template was used to mark the rows so that row spacings in all trays were exactly the same. This allowed placement of successive trays (and rows within each tray) in exactly the same wavebands for treatment.

In a typical experiment, Hendricks calibrated the spectrum on the treatment table and determined the wavelength and energy to be received at each row position. Kasperbauer determined row positions and placed the plants, and Borthwick manually opened and closed the shutter (door) on the spectrograph to obtain the desired irradiation time. After

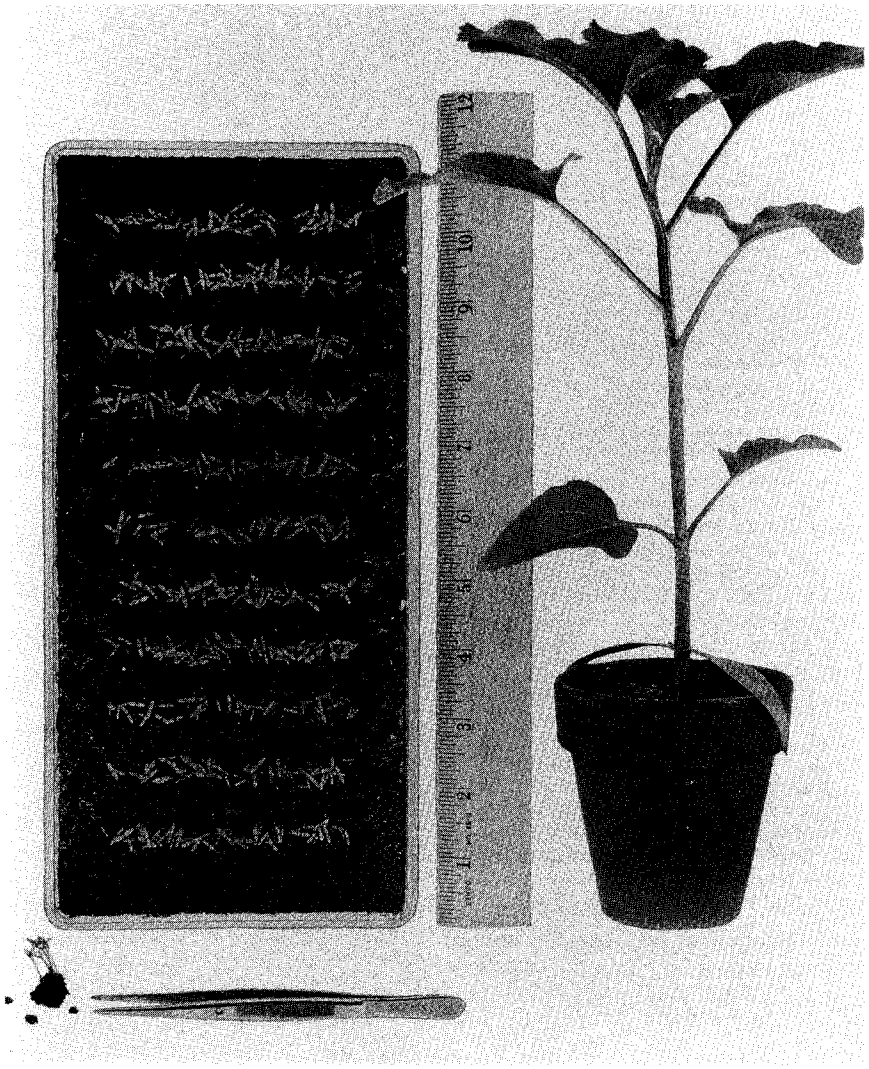

Figure 6 A tray of *Chenopodium rubrum* seedlings old enough for treatment on the spectrograph and a cocklebur plant also at an age suitable for treatment (units of measurement are in inches). The *Chenopodium* seedlings would be thinned to 10 per row, and the cocklebur trimmed of all but the most recently expanded leaf for treatment, as diagrammed for soybean in Figure 4.

five consecutive days of such treatment, the plants were grown in a noninductive greenhouse until the dark controls had visible florets. Then Kasperbauer placed all plants in a refrigerator to stop floral development, and examined and staged all plants. Floral development of each plant was assigned a numerical stage: 0.0 was completely vegetative and 9.0 indicated that florets were detectable without magnification (Fig. 7). Stages other than 9.0 were determined with the aid of dissecting needles and a magnifier. Floral stages (means for 10 plants) were plotted according to wavelength and energy received on the spectrograph to develop the action spectrum (efficiency of various wavelengths in control of floral development). The red action peak was at about 645 nm in these green seedlings (Fig. 8) and not at 660 nm as it is in light-requiring seed and in vitro. The shift from 660 to 645 nm was attributed to competitive absorption by chlorophyll at 660 nm in green plants (4). The treatment of red-irradiated plants in the far-red part of the spectrum showed that a few minutes at about 730 nm reversed the effect of red, but continued irradiation at that waveband caused a second reversal.

Treatment in the far-red part of the spectrum was done in two different ways: (a) following a saturation (7-min) exposure to red or (b) directly from darkness (Table 3). Exposure to far-red at about 735, 755, 775, and 795 nm took progressively longer to obtain a far-red effect (i.e., reversal of the inhibitory effect of a brief exposure to red light that was

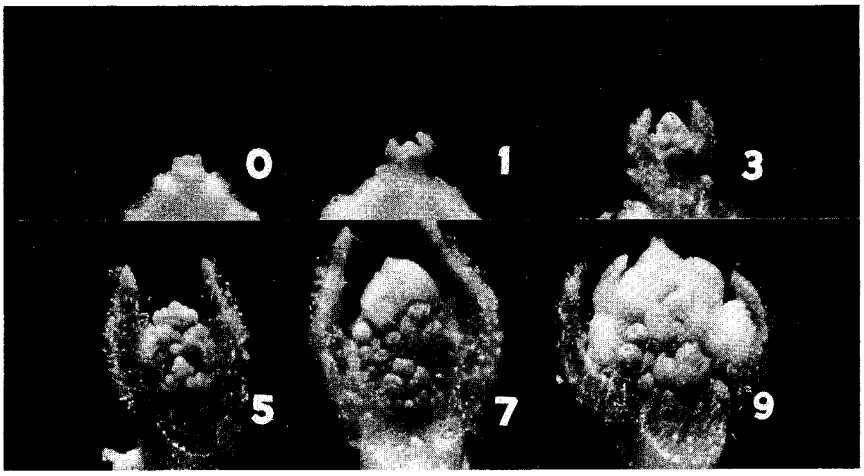

Figure 7 Floral stages of *Chenopodium rubrum* used to develop action spectra on the Beltsville spectrograph. (From Ref. 4.)

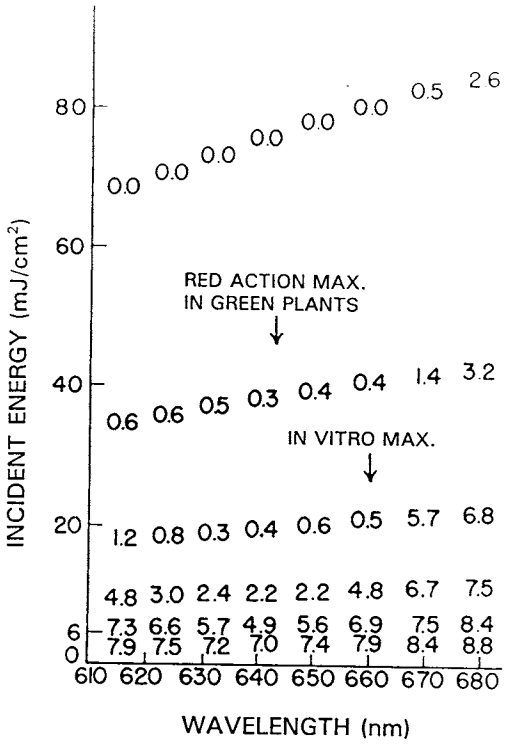

Figure 8 Red action spectrum for inhibition of flowering in *Chenopodium rubrum* seedlings irradiated at different wavelengths on the Beltsville spectrograph in the middle of the 16-hr night. Seedlings kept in darkness during these treatments (controls) attained stage 9.0. Each value is the average floral stage for 10 plants. (Source: Adapted from Ref. 4.)

applied just before placement of the plants in the spectrum). However, prolonged exposure to those wavelengths also produced progressively less red effect. The red effect of prolonged exposure to far-red was interpreted to be due to the overlapping of the Pr absorption curve into the far-red part of the spectrum and maintenance of a small but effective amount of phytochrome in the biologically active form long enough to inhibit floral induction (4). The different responses to far-red at 735 and 755 or even 775 nm (Table 3) later became a significant factor in interpreting plant responses to light reflected from other plants and from different colored soils, plant residues, and mulches (see a later section of this chapter). Similarly, the shifted red action peak from 660 nm (the in vitro absorption peak) to 645 nm in green plants (see Fig. 8) is also very important in the interpretation of plant responses under field conditions.

Table 3 Effects of Prolonged Irradiation at Selected Wavebands Immediately After (A) a Saturation Exposure to Red Light (Sufficient to Completely Inhibit Floral Development, i.e., Stage 0.0) or (B) Directly from Darkness (Dark Controls = Stage 9.0)

Waveband on spectrograph (nm)	Duration of irradiation (min)							
	0.5	1	2	4	8	16	32	64
A. After saturation exposure to red (controls = 0.0)								
645	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
735	2.8	6.8	7.3	7.5	7.7	7.2	5.9	1.1
755	0.7	6.3	7.1	7.4	8.0	8.2	6.3	5.2
775	0.1	1.7	5.7	7.2	8.1	8.1	6.7	6.3
795	0.0	0.0	0.4	3.1	6.8	7.2	6.8	6.3
B. Directly from darkness (controls = 9.0)								
645	2.6	0.3	0.1	0.0	0.0	0.0	0.0	0.0
735	9.0	9.0	9.0	8.9	8.7	8.0	6.6	0.6
755	9.0	9.0	9.0	9.0	8.8	8.4	8.0	7.1
775	9.0	9.0	9.0	9.0	9.0	8.7	8.3	7.6
795	9.0	9.0	9.0	9.0	9.0	8.9	8.7	8.2

Source: Adapted from Ref. 4.

Evidence obtained on the spectrograph and with narrow-band fixed filters suggested that immediately after the treatment with red light, the amount of the far-red–absorbing form of phytochrome (Pfr) would be high but would then decrease in amount while the amount of Pr would increase in darkness over a several hour period. Inhibition of flowering of short-day test plants was accomplished if the amount of Pfr was above a "critical level" for an "adequate duration." However, the critical level and adequate duration seemed to vary with plant species and stage of growth (15,16). Conversely, maintaining Pfr above a critical level for an adequate period resulted in promotion of flowering in long-day plants.

Since a brief irradiation with red light converted most of the phytochrome to Pfr and the proportion of phytochrome in the Pfr form diminished in darkness at a temperature-dependent rate, a series of short exposures to red light alternated with short dark periods could be almost as effective as continuous light for some plant species. Flowering of some short-day plants like *Chenopodium* would be inhibited by one brief exposure to red near the middle of the night, and repeated brief exposures to red did not further enhance that floral inhibition. However, chrysanthemum (*Chrysanthemum morifolium*) required longer exposures to light in order to completely inhibit flowering (17). Thus, chrysanthemum responded equally well to several hours of continuous

light or to short light–dark cycles during the same several hour period near the middle of the night (18,19). Flowering of long-day sweetclover plants also responded very little to a single brief exposure to light in the middle of the night. However, sweetclover (like chyrsanthemum) responded about the same to several hours of continuous light or to much less light applied for 10% of the time in short on–off cycles repeated over the same duration (20).

The spectrographic studies on photoconversion and timing of dark reversion (21) contributed to greenhouse studies with H. M. Cathey and Borthwick on cyclic lighting to regulate time of flowering. Cathey and Borthwick investigated short-day chrysanthemum (19) and Kasperbauer et al. the long-day sweetclover (20). Responses of long-day sweetclover plants to different cycle lengths are shown in Figure 9. Cycle length could be less frequent with light from cool-white fluorescent lamps than from incandescent filament lamps because these sources differed in the far-red/red ratio in the light that they emitted. The fluorescent lamps emitted very little far-red. Consequently, light from fluorescent lamps put a greater fraction of phytochrome in the Pfr form and more time could elapse before the Pfr level dropped below the "critical" level for control of flowering. While fluorescent lamps were more efficient in cycle length, incandescent filament flood lamps were more convenient and equally effective if the cycle length was shorter. Cyclic lighting effectively controlled time of flowering with only 10–20% of the electrical energy that is needed for continuous lighting. During recent years, cyclic lighting has been used to make the mountains of the tropics a desirable place to produce cut flowers commercially all year. The natural day lengths are about the same all year near the equator, and temperatures differ with elevation. With these natural background conditions, cyclic lighting can be used to delay floral induction of short-day plants until they attain suitable size, after which they can be brought to flower under the natural length days. On the other hand, long-day plants such as carnation can be kept vegetative on the natural days until they are ready to be placed under cyclic lighting to induce flowering. It is obvious that basic studies on the photoconversion of phytochrome and timing of dark reversion were important aspects of the real world use of this information.

Controlled Environment Studies

The use of prolonged exposures (up to 90 min) to far-red light, and the energy requirements for photoconversion and the timing of dark reversion of phytochrome (4,21) in regulation of a physiological process, flowering, provided the foundation for many controlled-environment and

Figure 9 Sweetclover (long-day) plants grown under 8 hr of sunlight and supplemental incandescent filament light applied during the 16-hr night as follows (left to right): 16 hr continuous, 1.5 min every 15 min, 6 min every 60 min, 24 min every 4 hr, 96 min centered at midnight, and no supplemental light. Plants were 3 months old when treatments began. Photographs were taken after 6 weeks of the daily treatment. (From Ref. 20.)

field studies. There are numerous excellent examples of phytochrome regulation of plant physiological processes under controlled environments in many laboratories around the world. However, because of space limitations this section will be confined to some selected experiments with intact green plants that led to a better understanding of phytochrome action in field growth and development of crop plants. Many of the examples are with tobacco, the same species that launched the scientific curiosity of Garner and Allard (1) and led to their discovery of photoperiodism.

The controlled environment experiments discussed herein were started with tobacco in the early 1960s. They involved determination of phytochrome regulation of leaf shape and thickness, internode length, chlorophyll concentration, chlorophyll a/b ratios, chloroplast structure, photosynthetic efficiency of leaves, and accumulation of compounds such as sugars, starch, epicuticular alkanes, fatty acids, amino acids, organic acids, alkaloids, and polyphenolics. The objective of the controlled-environment experiments was to learn how and why plants responded as they did to light variables.

For most of these studies, plants within a given experiment were grown under identical conditions in the same controlled environment for about 23 hr and 50 min each day. At the end of the bright light period each day, plants were moved to adjacent rooms and irradiated with either 5 min of red, 5 min of far-red, or 5 min of far-red followed immediately by 5 min of red light to test for photoreversible control. This approach allowed study of phytochrome regulation of developmental processes when temperature and photosynthetic light were kept constant among all plants. The red light put most of the phytochrome in the Pfr form whereas the far-red put most of the phytochrome in the Pr form at the beginning of the night. This allowed the phytochrome form to initiate physiological events during the night that regulated how the plants invested (partitioned) the photosynthate that had accumulated at the end of the photosynthetic period. The working hypothesis was that responsiveness to red and far-red was related to adaptation of plants to various environments in the real world. Some of the controlled-environment responses that became highly relevant in the interpretation of field plant responses and their management are summarized below.

Tobacco plants that received a brief exposure to far-red at the end of each day developed stem and leaf characteristics that were dramatically different from those that received a brief exposure to red at that time. Also, plants that received 5 min of far-red followed immediately by 5 min of red responded to the kind of light received last. This photoreversible regulation of developmental responses was evidence that phytochrome was involved in initiating physiological events in plant develop-

ment. Since phytochrome action was shown to be dependent on the photoequilibrium between the two forms of phytochrome (21), it was reasonable to think of end-of-day far-red as either a low red/far-red ratio or as a high far-red/red ratio. Because responses to far-red were very dramatic in growing seedlings and there is much competitive absorption of red at 660 nm (the phytochrome absorption peak) in green plants, far-red was projected as the more important variable in nature (22). Thus, this author uses the far-red/red ratio. This concept is consistent with a study by Vogelmann and Bjorn (23) who inserted fiber optic probes into fleshy leaves to compare the amount of far-red light that reached different depths within the leaf tissue relative to the amount received at the exterior surface. They detected higher amounts of far-red (at 750 nm) inside the fleshy leaves, which they attributed to photon scattering within tissue that had relatively little competitive absorption of far-red light.

Leaves that developed on plants that received far-red (a high far-red to red ratio) at the end of the daily photosynthetic period grew longer and narrower than those that received a low ratio at the end of each day (Fig. 10). The petioles were longer as were the stem internodes (Fig. 11).

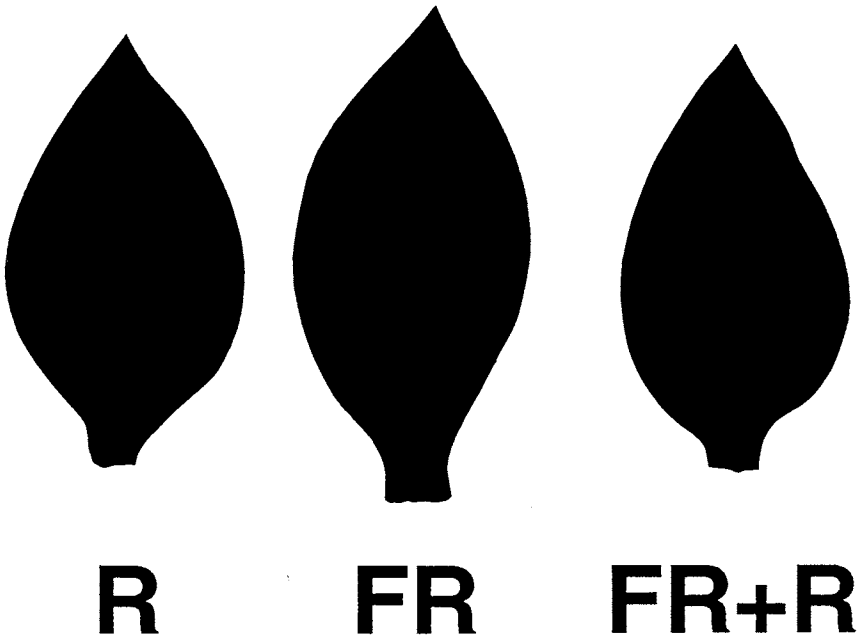

Figure 10 Tobacco leaves from plants that received 5 min of red (R), 5 min of far-red (FR), and 5 min of far-red followed immediately by 5 min of red (FR + R) at the end of each day during development. (Adapted from Ref. 24.)

Figure 11 Tobacco plants that received (from left to right, respectively) 5 min of red, 5 min of far-red, or 5 min of far-red followed immediately by 5 min of red at the end of each day for 21 days.

Other plant species such as soybean responded similarly. As with tobacco, biomass partitioning among leaves, stem, and roots in soybean seedlings was highly influenced by the far-red/red light ratio received just before darkness (see Table 4). In addition to the differences in leaf shape (Fig. 10), leaves that received the higher far-red/red ratios were thinner, had a higher chlorophyll a/b ratio (24), and a higher concentration of light-harvesting chlorophyll protein (LHC-II) (26). Chloroplasts in leaves that developed with the higher far-red/red ratio (far-red treatment) had more but smaller grana and smaller starch grains (27). Far-red–treated

Table 4 Effects of End-of-Day Red (R) or Far-Red (Low and High FR/R Ratio, Respectively) on Percentages of Dry Biomass Partitioned to Leaves, Stems, and Roots of Soybean Seedlings Under Controlled Environments

End-of-day		Dry biomass % in:			
Light[a]	FR/R ratio	Leaf blades	Stems and petioles	Roots	Shoot/root ratio
R	Low	43.9	23.6	32.5	2.1
FR	High	43.6	33.2	23.2	3.3
FR, R	High, low	43.4	22.8	33.8	2.0

[a] R and FR treatments were for 5 min at the end of each day for 20 consecutive days. The FR, R treatment received 5 min FR followed immediately by 5 min R each day.
Source: Adapted from Ref. 25.

leaves had higher concentrations of sugar and organic acids, and lower concentrations of amino acids (Table 5). In addition to being thinner with higher chlorophyll a/b ratios, the leaves that developed on plants that received the higher far-red/red ratio were more efficient photosynthetically (29). That is, they fixed more CO_2 per mass of leaf tissue (Fig. 12) even though they did not differ on a leaf area basis. Those combined observations in controlled environments suggested that the amount of far-red and the far-red/red ratio played a major role in development of plant characteristics that could favor survival while competing with other plants. It was apparent that the amount of phytochrome in the Pfr form relative to the total amount of phytochrome (P), particularly at the beginning of a dark period, plays a critical role in signaling photosynthate distribution and developmental patterns.

It was not clear, however, whether a low Pfr/P ratio (the consequence of irradiation with far-red) triggers a chain of metabolic events leading to "competition-adapted" development or whether the events occur because the Pfr/P ratio is too low to signal a chain of events leading

Table 5 Concentrations of Free Sugars, Organic Acids, and Amino Acids in Tobacco Plants that Received 5-min Far-Red or 5-min Red (High or Low FR/R Ratio, Respectively) at the End of Each Day During Development

	End-of-day radiation and plant part					
	FR (high FR/R)			R (low FR/R)		
Component	Leaf blade	Mid-rib	Stem	Leaf blade	Mid-rib	Stem
Free sugars (mg/g dry matter)						
Sucrose	7.5	8.3	12.5	6.3	7.5	17.5
Glucose	3.0	19.2	45.0	1.5	2.7	10.0
Fructose	2.8	10.3	40.0	2.0	1.3	10.1
(Total)	(13.3)	(37.8)	(97.5)	(9.8)	(11.5)	(37.6)
Organic acids (mg/g dry matter)						
Malic	16.3	50.1	13.0	12.5	50.1	12.5
Citric	2.7	1.5	<0.5	2.5	1.4	<0.5
Succinic	3.0	3.0	4.0	2.5	3.0	3.8
Fumaric	1.3	2.5	<0.5	0.5	2.5	<0.5
Ascorbic	1.8	6.3	3.0	1.5	6.3	2.8
(Total)	(25.1)	(63.4)	(21.0)	(19.5)	(63.3)	(20.1)
Free amino acid (μM/g dry matter)						
(Total)	(44.2)	—	—	(66.4)	—	—

Source: Adapted from Ref. 28.

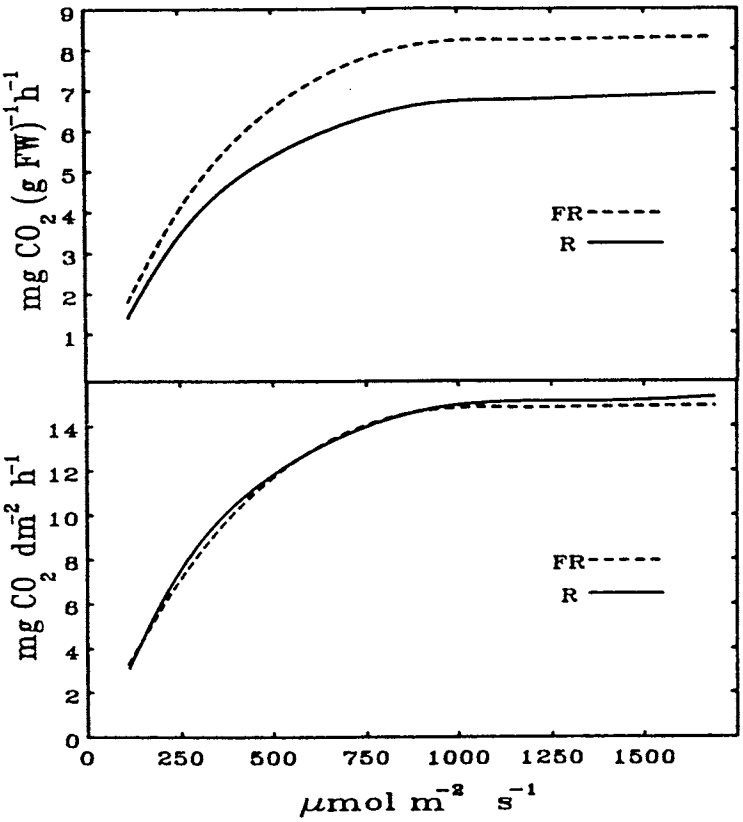

Figure 12 Net CO_2 assimilation rates of tobacco leaves that received 5 min of red (R, a low FR/R ratio) or far-red (FR, a high FR/R ratio) at the end of each day during development. Data are expressed on the basis of fresh weight of leaf lamina, and leaf area. (Curves are drawn from Ref. 29.)

to "sun-adapted" development. Whichever the case, some effects of far-red are similar to those of exogenous gibberellic acid (GA). The thin leaves, light color, and somewhat elongated internodes of plants treated with GA (30–32) and end-of-day far-red (29) suggested that both treatments may involve the same metabolic pathway. Both GA- and far-red–treated plants have decreased total chlorophyll, and the reduction of chlorophyll b is greater than the reduction of chlorophyll a resulting in an altered chlorophyll a/b ratio. It was suggested that end-of-day far-red, through its influence via the phytochrome system during the dark period, may initiate shifts in the balance of naturally occurring growth regula-

tors such that the imbalance tips in favor of gibberellins much the same as when exogenous GA is added to a plant.

Free amino acid concentrations in tobacco leaves also differed with photoperiod as well as with the far-red/red light ratio received at the end of each day (Table 6). Wilkinson hypothesized that photoperiod and plant competition for light are sufficient to alter endogenous GA and enzyme systems that regulate metabolic pathways and endproducts in a field environment (34). Epicuticular alkanes differed significantly between two ages of leaves from the same field-grown tobacco plants (35). Since the field-grown leaves developed at different times under changing natural environments, plants of a genetically uniform line of tobacco were grown under different photoperiods in controlled environments. Epicuticular alkanes, fatty acids, and fatty alcohols were all highly influenced by growth environment (Table 7). Other investigators (37) have shown that exogenous [^{14}C]-fatty acids could be incorporated into intracellular lipids. Also, when $^{14}CO_2$ was applied to leaves with or without GA_3 treatment, there was a greater translocation of ^{14}C from the GA-treated leaves (38) even though the GA treatment did not result in differences in total ^{14}C fixed in the leaves. And synthesis of some enzymes has been induced by GA_3 (39) whose synthesis is photoperiodically controlled (40). Modified epicuticular content and composition associated with light parameters are examples of metabolic alterations in response to environmental changes that may increase the probability of survival.

Table 6 Free Amino Acid Content of Tobacco Leaves that Developed at 25°C Under 8- or 16-hr Daily Light Periods that Ended With 5 min Red (R, Low FR/R Ratio) or 5 min Far-red (FR, High FR/R Ratio) Each Day

	End-of-day		Amino acid group		
Photoperiod	Light	FR/R ratio	Oxalacetate	α-Ketoglutarate	Pyruvate
			μM/g dry wt[a]		
Short	R	Low	24 a	15 a	13 a
	FR	High	14 c	13 b	10 b
Long	R	Low	14 c	10 c	8 c
	FR	High	17 b	8 d	6 d

[a] Values are means for 15 plants. Within each column, values followed by the same letter do not differ significantly at the 5% level.

Source: Adapted from Ref. 33.

Table 7 Epicuticular Fatty Acid, Fatty Alcohol, and Alkane Concentrations in Tobacco Leaves Grown in Controlled Environments Under Short or Long Photoperiods at 28°C

			Alkanes (n)			
Photoperiod	Fatty acids	Fatty alcohols	C_{27}	C_{29}	C_{31}	C_{33}
	ng/cm^2		$\mu g/cm^2$			
Short	1306	1524	12	40	266	221
Long	2549	840	45	50	586	710
Signif.	*	*	*	*	*	*

* Indicates that differences are statistically significant at the 5% level of confidence.
Source: Adapted from Refs. 35 and 36.

The controlled-environment studies suggested that the ratio of far-red relative to red photons received by developing leaves of a field-grown plant could influence adaptive morphological development of the plant and photosynthetic efficiency of the leaves. An interesting analogy would be to think of the phytochrome system within the growing plant as a variable sensor that is constantly monitoring the far-red/red ratio as an indicator of competition from other plants and as an initiator of physiological events that favor survival of the plant among that perceived competition. For example, a higher far-red/red ratio leads to a longer stem with fewer branches and a more efficient photosynthetic system. This adaptive response would favor survival by increasing the probability of keeping some leaves in sunlight above competing plants and perhaps by having leaves that are more efficient in utilizing light within the plant canopy. It was evident from the controlled-environment studies that the phytochrome system can initiate physiological events leading to adaptive morphological development such that the plant is better suited to compete with other plants in its growth environment, and that genetically identical plants can differ significantly in quantities of various chemical constituents, depending on growth environment.

Field Plant Response to Far-Red Reflected from Other Plants

This author's field plant population density studies were started with tobacco in the mid-1960s and extended to other species in the early 1980s. The initial field observations of end vs. mid-row tobacco plants showed that the mid-row plants were taller, had slightly longer internodes and slightly thinner leaves. These characteristics were in the direction of controlled-environment plants that received the higher far-red/red ratios.

Growth of pretransplant tobacco seedlings followed the same pattern. That is, close-spaced seedlings had longer and thinner stems, narrower leaves, and a smaller root system relative to seedlings that were wider apart. The characteristics began to appear even before mutual shading occurred among the close-spaced seedlings. These outdoor observations combined with controlled-environment observations strongly suggested that the far-red/red ratio and phytochrome might be involved in tobacco response to nearness of other plants. The "tools" available to study field population effects on light spectra were somewhat primitive in the 1960s; however, critical measurements were made. The available portable spectroradiometer could measure only 16 fixed wavebands. Of these, 11 were between 390 and 700 nm, one was at 725 nm, and another at 791 nm. The light detector head contained four rows of four fixed filters, each 2.5 cm × 2.5 cm, and there was a manual switch to change wavebands. Thus, it was cumbersome by today's standards. However, the detector head was on a 3-m cable and it was possible to measure transmission through a single tobacco leaf and at various places within the grown canopy (22). The data showed that green leaves absorbed most of the blue and red, but transmitted some of the green and much of the far-red. These measurements also revealed that a higher percentage of light was transmitted at 791 than at 725 nm. The measurements documented differences in spectral balance of light within and below a plant canopy (Table 8). However, the differences in far-red/red ratios within and below the tobacco canopies could not explain why the upper leaves (those in sunlight above competing leaves) were longer, narrower, and thinner when plants were grown close together. These observations suggested (see Figs. 10 and 11) that the upper leaves in close-spaced tobacco received more far-red or at least a higher far-red/red ratio than upper leaves on an isolated plant. As part of the measurements and comparisons, light at each measured waveband in the canopy was expressed as a percentage of the incoming sunlight at the same waveband. An observation that began as a puzzle in the late 1960s became the critical factor in relating field plant population density to phytochrome-regulated adaptive morphological development.

When light measurements at various points within and below the tobacco canopy were compared with incoming sunlight, some dramatic differences were observed at the 791-nm waveband depending on whether the incoming light was measured on a road away from other plants or in a small patch of sunlight on the ground within the tobacco fields (see footnote, Table 8). Subsequent comparison of the spectra of light on the roadway with that on the soil surface in the tobacco field showed that the light close to plants had more far-red than that on the

Table 8 Percentages of Incoming Sunlight Received Within and Below a Canopy of 190-cm-tall Tobacco at About 1 p.m. on September 1, 1967 near Lexington, KY

Peak wavelength (nm)	Percentage of incoming sunlight[a] detected:		
	Below a single leaf	Within canopy	Below canopy
391	1.7	0.9	0.5
432	0.5	0.7	0.3
448	0.7	0.7	0.3
483	0.9	0.6	0.4
511	3.3	0.8	0.6
543	22.7	11.0	6.5
576	14.7	5.0	3.4
601	10.8	2.6	2.1
629	7.9	1.7	1.4
658	6.1	2.3	1.7
686	6.6	2.2	1.9
725	27.5	11.6	8.8
791	49.5	36.3	20.3

[a] The incoming sunlight was measured on a road, away from tall plants.
Note: Light at 791 nm was about 15% higher in sunflecks on the ground near tobacco plants than it was above the road, away from tall plants.
Source: Adapted from Ref. 22.

roadway. The only logical explanation was that the extra far-red in the tobacco field was reflected from the nearby plants. Far-red reflection from other plants also helped explain why the upper (unshaded) leaves of close-spaced tobacco had a different shape than upper leaves on isolated plants. Similarly, the stem length and leaf shape differences of close-spaced and isolated tobacco pretransplant seedlings could be explained when far-red reflected from other seedlings was considered as a contributor to the far-red/red ratio received by the growing plants.

Beginning in the early 1970s tobacco plants were routinely set at three population densities to study plant morphological development and leaf chemistry. These studies were highly relevant as a background for possible alternate production procedures, and also as a test of plant spacing, the far-red/red ratio, and the resultant physical and chemical characteristics of plants as related to closeness of other plants. Close-spaced plants were usually 30 × 30 cm, normal spacing was 45 cm apart in rows that were 100 cm apart, and wide-spaced plants were 120 × 120 cm apart. Although precise spectral measurements were difficult with the

available portable spectroradiometer, higher far-red levels were detected at a midpoint between the close-spaced plants than at the mid-point between wide-spaced plants. That is, the detected level of far-red was higher when the spectroradiometer light collector was closer to a growing plant.

The physical and chemical effects of spacing on the plants were more easily documented. Close-spaced plants began growing taller than wide-spaced plants even before mutual shading began. Representative plants taken from the field 6 weeks after transplanting are shown in Figure 13. Notice that close-spaced plants were taller (longer internodes), with thinner stems and narrower leaves. All of these features were consistent with those associated with a higher far-red/red ratio in the controlled-environment studies (see Figs. 10 and 11). In addition to being taller with narrower leaves, the close-spaced plants had thinner leaves, higher chlorophyll a/b ratios, lower alkaloid concentrations, and higher chlorogenic acid concentrations (Table 9). These plant responses followed the same trends as plants that received higher far-red/red ratios in the controlled-environment studies (Table 10).

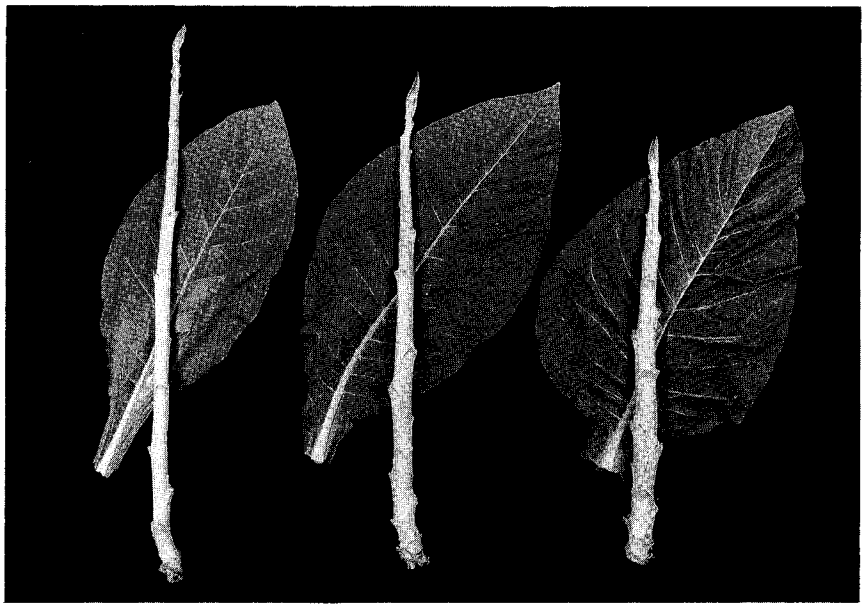

Figure 13 Tobacco plants after 6 weeks of growth in (from left to right, respectively) close, normal, and wide spacings in a field.

Table 9 Effects of Field Plant Spacing on Concentrations of Alkaloids and Phenolics (Chlorogenic Acids) in Mature Tobacco Leaves

Component	Plant spacing during growth		
	Close	Normal	Wide
		(mg/g)	
Total alkaloids	13 c*	32 b	54 a
Chlorogenic acid	0.57 a	0.32 b	<0.05 c

*Values in the same line that are followed by the same letter do not differ significantly at the 5% level.
Note: The close-spaced plants received higher FR/R ratios during growth.
Source: Adapted from Ref. 41.

In 1983, we extended the studies to other plant species and obtained a much improved portable spectroradiometer that was capable of measuring radiation from 300 to 1100 nm at 2-nm intervals. With a remote light collector on a 1.5-m fiber optic probe, we measured the light spectra received at the upper surface of soybean canopies in north–south vs. east–west rows and in various other spacing combinations. Light measurements were taken near the tops of growing plants because Parker and Borthwick (43) had shown that the most recent fully expanded leaves were very efficient in sensing morphogenic light signals that regulate developmental responses in the growing parts of the plant.

Table 10 Concentrations of Alkaloids and Phenolics in Leaves of Tobacco Seedlings that Received 5 min of Far-Red (FR, High FR/R Ratio) or 5 Min Red (R, Low FR/R Ratio) at the End of Each Day in Controlled Environments for 18 Days

Component	End-of-day radiation and plant part			
	FR (high FR/R ratio)		R (low FR/R ratio)	
	Blades	Mid-ribs	Blades	Mid-ribs
		(mg/g dry matter)		
Alkaloids				
Total	3.9	0.5	7.0	0.8
Phenolics				
Chlorogenic acid	20.0	10.0	18.8	6.3
Total	24.0	20.3	22.1	13.1

Source: Adapted from Ref. 42.

The absorption, reflection, and transmission spectra were also determined from individual leaves of a number of species, including soybean. Representative spectra from soybean leaves are shown in Figure 14. It was clearly evident that each green leaf reflected almost half of the far-red that reached it and that the reflection "plateau" begins at about 750 nm. Thus, it was reasonable to expect that the number, nearness, and size of competing plants would influence the amount of reflected far-red and the far-red/red ratio received by a nearby growing plant. Also, it was considered possible that row direction might further influence the far-red/red ratio received because of heliotropic movement of leaves, causing them to be directional far-red reflectors. Further, it is important to note that the far-red/red ratio in incoming sunlight also increases near sundown.

Clearly, the far-red/red ratio at the surface of the upper leaves was largely affected by the amount of far-red reflected from nearby plants (44). The ratio was also influenced by heliotropic movement of leaves of the broad-leaf, long-petiole soybean plants, which had the effect of directional far-red reflectors, especially near the end of the day. Thus, it was

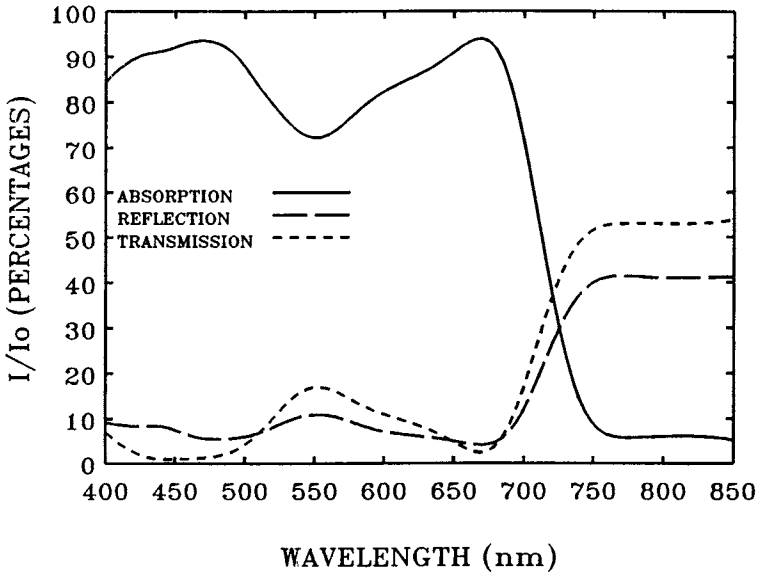

Figure 14 Light absorption, reflection, and transmission spectra from a soybean leaf. The absorption percentages were calculated by subtracting the combined values for reflection plus transmission from 100 at each measured wavelength. (Redrawn from Ref. 25.)

hypothesized that incoming sunlight with a higher far-red/red ratio that is reflected off of the heliotropic leaves could contribute to an important end-of-day "signal" of the relative amount of competition from other plants, and this might be influenced by row direction in broad-leaf, long-petiole plants.

Incoming light from all four directions was measured near the tops of bean plants growing in north–south vs. east–west rows at different stages of growth. When numerous readings taken throughout the day were averaged, plants in north–south rows received higher far-red/red ratios. An example is shown in Table 11. As predicted, this pattern of higher far-red/red ratio in north–south rows was most evident near the end of the day when the heliotropic movement of the leaves had the effect of reflecting light back to the adjacent row.

As projected from the earlier controlled-environment experiments, a higher amount of reflected far-red and the associated higher far-red/red ratio resulted in slightly taller shoots and less branching of soybean seedlings in north–south vs. east–west rows (Table 12). There were several examples in which the larger shoots produced more seed or fruit, when there was no moisture or soil nutrient stress (25,45).

In a wheat (*Triticum aestivum* L.) experiment with Karlen (46), we measured light spectra at the soil surface within different populations of field-grown seedlings in early spring (Fig. 15). As expected, close-spaced plants received more reflected far-red and higher far-red/red ratios. The

Table 11 Photosynthetic and Photomorphogenic Light Received at the Shoot Apex of Bush Bean Plants Grown in North–South (N-S) vs. East–West (E-W) Rows, and Plant Productivity

Characteristic	Row orientation	
	N-S	E-W
Light (means of 24 readings)[a]		
Photosynthetic (μmol/m^{2}s)	389 ± 62	393 ± 59
Photomorphogenic (FR/R photon ratio)	1.85 ± 0.23	1.48 ± 0.13
Plant productivity		
Green beans (g fresh wt/plant)	59.0 ± 8.3	43.0 ± 4.1

[a] Light coming to the shoot of two representative plants from each row orientation was measured from the north, south, east, and west at 11:00 a.m., 1:30 p.m., and 3:30 p.m. on a cloudless day near Frankfort, KY. Each light value in the table is the mean ± SE for the two plants, four directions, and three times during the day (i.e., means are for 24 separate readings).

Source: Adapted from Ref. 45.

Table 12 Row Orientation Effects on Characteristics of Soybean Plants Grown in North–South (N-S) vs. East–West (E-W) rows in Irrigated Loamy Sand in Field Plots near Florence, SC

	Row orientation	
Characteristic	N-S	E-W
At 6 weeks (means/plant)		
Stem length (mm)	348 ± 6	324 ± 5
Nodes/stem (no.)	8.1 ± 0.1	8.2 ± 0.1
Branches/plant (no.)	1.8 ± 0.4	3.0 ± 0.2
At harvest (dry matter/1-m row)		
Seed (g)	158.2 ± 7.8	142.8 ± 7.5
Pods (g)	58.2 ± 3.0	53.0 ± 4.0
Stem (g)	43.8 ± 1.9	40.8 ± 3.2
Seed/straw (ratio)	1.55	1.52

Source: Adapted from Ref. 25.

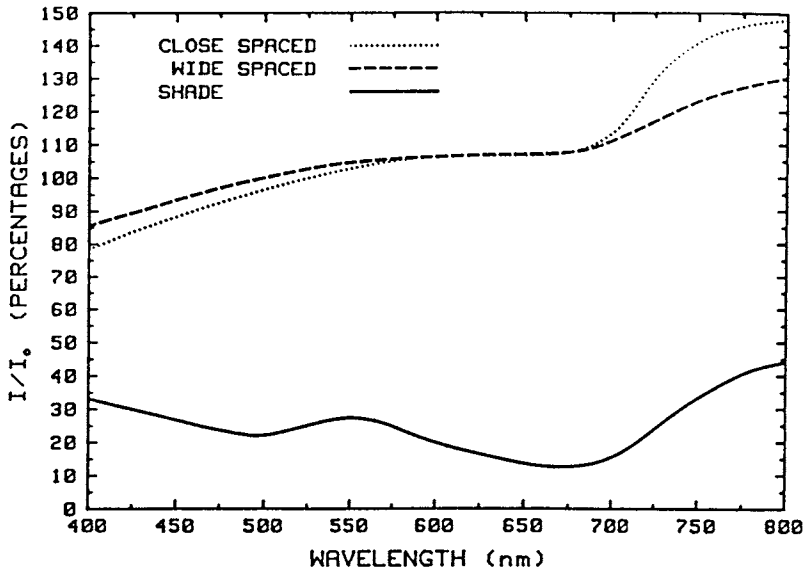

Figure 15 Spectral distribution of light in shade (solid line) and in sunflecks (dashed lines) at soil level in a field of close-spaced and wide-spaced wheat seedlings (about 8–10 cm tall) near Florence, SC in early afternoon in mid-March 1984. Values are expressed as percentages of incoming sunlight at each measured wavelength. (From Ref. 46.)

plant response was to develop fewer tillers (analogous to fewer branches on soybean seedlings) and longer leaves, again showing a phytochrome response to reflected far-red under field conditions. This was an example of how the phytochrome system within the wheat seedlings could sense (measure) the nearness of competing plants and then initiate physiological events that regulated the amount of tillering. That is, when soil moisture and nutrients were not limiting, the phytochrome system measured the far-red/red ratio, sensed the amount of competition, and regulated the amount of tiller development. Representative wheat seedlings from wide- and close-spaced plantings are shown in Figure 16.

There were a number of other population and spacing studies, some of which involved the use of plants in containers imbedded in soil among the various populations. This allowed determination of effects of spacing and the far-red/red ratio on shoot-root biomass ratios as a result of phytochrome regulation of partitioning within a growing plant, when the container-grown plants in all population densities had the same volume of the same soil mixture. Again, as projected from controlled-environment studies (see Table 4), plants that were closer together and received the higher far-red/red ratios had the higher shoot/root biomass ratios. Plants that received the higher far-red/red ratios in the field as well as those in controlled environments partitioned higher percentages of the new photosynthate to growing stems, and less to branches (or tillers) or new root growth. This is a reasonable adaptive response because the far-red/red ratio would be sensed as an indicator of competition, and plants with longer stems would have a greater probability of keeping some leaves in sunlight above competitors and surviving long enough to produce the next generation.

Upwardly Reflected Light from Soil Surfaces

When it was clear that plants respond to spectral composition of light reflected from other living plants, P. G. Hunt (a soil scientist) and I decided to find out whether plants would respond morphologically to spectral differences in light reflected from different colored soils, plant residues, or other soil covers (mulches). In the initial studies during 1984 and 1985, the spectra of upwardly reflected light were measured 10-cm above five different colors of soil (47). This height above soil was selected because it is in the seedling establishment zone, and seedlings are extremely responsive to spectral composition of light (4,21). Measurements were made over dry or wet soil and over the soils when they were about 80% covered with plant residue from a previous crop. The different colored soils and plant residues reflected different far-red/red ratios.

Figure 16 Field-grown wheat seedlings (about 8 to 10 cm tall) from wide-spaced (left) and close-spaced (right) population densities in mid-March 1984. (From Ref. 46.)

The next step was to determine whether soil surface color could influence reflected light sufficiently to modify seedling growth. Soybean seedlings were started in pots of soil and placed on greenhouse benches about 60 cm apart in groups of four. Each group of four soybean seedlings was covered with a 122 × 122 × 2 cm insulation panel that had four 2.5-cm holes 60 cm apart so that the seedlings could grow through the insulation panels. The panels were covered with about 5 mm of the different colored soils, or soil that was about 80% covered with straw. In this manner, root temperature differences were minimized below the different soil surface colors. Plants over the brick-red soil and over the straw residue received higher reflected far-red/red light ratios than those grown over the white soil (47,48); and they grew taller, had less root growth, and developed higher shoot/root biomass ratios (48). These initial studies were very significant because soils are of many colors as are plant residues left on the soil surface in many no-tillage or other conservation tillage procedures.

Other experiments were done in which the insulation panel surfaces were painted instead of being covered with different colored soils or plant residues. Plants responded the same to either painted or soil-covered surfaces if they reflected the same spectrum of light. That is, plants grown over the red painted surfaces received higher reflected far-red/red ratios than plants grown over white, and they grew taller and had higher shoot/root biomass ratios. Subsequently, painted surfaces were used for outdoor experiments because soils and plant residues were affected by wind and rain. Clearly, soil surface color could affect the reflected far-red/red light ratio sufficiently to influence photosynthate partitioning and biomass distribution within growing seedlings.

In late 1985, D. R. Decoteau (a horticulturist) observed the soybean seedling responses to light reflected from different colors of painted or soil-covered surfaces (described above). He proposed that the concept be extended to irrigated field-grown tomatoes (*Lycopersicon esculentum*) for the 1986 season. Since black or white plastic mulches were widely used for soil and water conservation as well as to control weeds in the production of tomato and other high-value food crops, we then explored the possibility that an altered surface color on the mulch could maintain those benefits and have an added favorable affect on plant productivity. The working hypothesis (based on many previous experiments that involved controlled environments, reflection from other plants, and reflection from colored soils, plant residues, and colored panels) was that an upwardly reflected far-red/red ratio higher than that in incoming sunlight would signal the plant to partition more of the new photosynthate to shoots, whereas a lower ratio would favor partitioning to roots. Irri-

gated tomatoes grown in sunlight over mulches with red surfaces produced significantly increased fruit yield relative to those grown with conventional black or white mulches (49). This response was consistent with the hypothesis, i.e., the red paint used to change the surface color of the plastic mulch reflected a higher far-red/red ratio, and the tomato plants partitioned more photoassimilate to shoots, including fruit. Subsequent experiments with a wide range of colored mulches and a number of mulching materials and plant species have confirmed that the spectrum (particularly the far-red/red ratio and the quantity of blue) of upwardly reflected light over colored soil surface covers (mulches) can regulate fruit number and size, leaf shape and thickness, concentrations of chlorophyll and light-harvesting chlorophyll protein, root size of turnip, and even the yield of cotton (26,50).

SUMMARY

Photomorphogenesis plays a very important role in utilization of photosynthate within the growing plant. It is important to realize that the strategy of each plant is to survive long enough in its existing environment to produce the next generation. Thus, the plant must be able to sense the total environment, integrate the information, and adapt to the constantly changing environmental conditions. Examples presented in this chapter involved the light environment primarily as it is affected by season and competition from other plants. The phytochrome system within the growing plant functions as a constant sensor of photoperiod and competition from other plants, and then regulates initiation of metabolic events that result in adaptive responses such as stem length, leaf shape and thickness, leaf waxes, amount of branching (or tillering), relative root size, and flowering.

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