PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Perception of Shade [and Discussion]

M. G. Holmes, A. M. Farmer and M. R. Bartley

Phil. Trans. R. Soc. Lond. B 1983 **303**, 503-521
doi: 10.1098/rstb.1983.0110

Phil. Trans. R. Soc. Lond. B **303**, 503–521 (1983)
Printed in Great Britain

Perception of shade

By M. G. Holmes

Radiation Biology Laboratory, Smithsonian Institution, 12441 Parklawn Drive, Rockville, Maryland 20852, U.S.A.

Plants perceive shade by responding to both the fluence rate and to the spectral quality of the natural radiation environment. Changes in fluence rate are perceived by separate photoreceptors absorbing in both the blue and the red wavebands. The identity of the photoreceptor (or photoreceptors) responding to changes in the fluence rate of blue light is unknown (see Briggs, this volume). Physiological responses to changes in the fluence rate in the red waveband appear to be mediated through phytochrome. The relative roles played by the blue-light-absorbing photoreceptor and phytochrome in determining the response to changes in fluence rate varies between species and organs and is also dependent on the physiological age of the plant. Evidence is also presented that supports the concept that phytochrome functions to perceive the specific form of shade caused by surrounding competitive vegetation.

1. Introduction

To perceive shade, a plant must possess one or more photoreceptors that react to changes in the natural light environment. The reaction to a light stimulus provides a means by which information on the prevailing light régime can be transduced in a form which enables, or causes, appropriate physiological responses. With this approach it is inevitable that energy-transducing photoreceptors, such as chlorophyll, are excluded from the discussion and that the study centres on signal-transducing photoreceptors that may enable a plant to react appropriately to shade. The method used here is to compare the spectral properties of natural light with the known properties of signal-transducing photoreceptors.

2. Shading by vegetation

Plants growing within vegetation shade experience a greatly different radiation environment from those growing in open habitats. Green leaves absorb, reflect and transmit light selectively. Blue and red wavebands are absorbed strongly, green light is absorbed less strongly and far-red radiation is largely reflected or transmitted. These patterns of selective absorption and scattering have also been observed under vegetation canopies (see, for example, Federer & Tanner 1966; Vezina & Boulter 1966; Stoutjesdijk 1972*a*, *b*; Holmes & Smith 1975; Holmes & McCartney 1976; Holmes 1981). The attenuating effects of a wheat (*Triticum aestivum* L.) canopy on natural daylight are shown in figure 1. Three spectral changes in daylight as it passes through the canopy are of known physiological significance. First, the quantity of photosynthetically active radiation (p.a.r.) (400–700 nm) is strongly reduced. This reduction can affect dry mass accumulation rate. Second, the reduced quantity of blue radiation may be of importance to the fluence-rate-dependent responses controlled by a blue-light-absorbing photoreceptor. The third spectral change that is known to be of physiological importance is

the strong depletion of the red waveband and relatively weak depletion of the far-red waveband. This spectral change is detected by phytochrome.

3. Perception of light quality

To detect light quality, a plant must be able to compare the relative quantities of light in two or more wavebands (Smith 1981*a*). This can be achieved by two methods. One possibility is the interaction of two separate photoreceptors whose mediated response depends on the relative amounts of light absorbed by each photoreceptor. There are several responses known to be controlled by two separate photoreceptors, but quantitative information on how the two photoreceptors interact is limited (Mohr 1980).

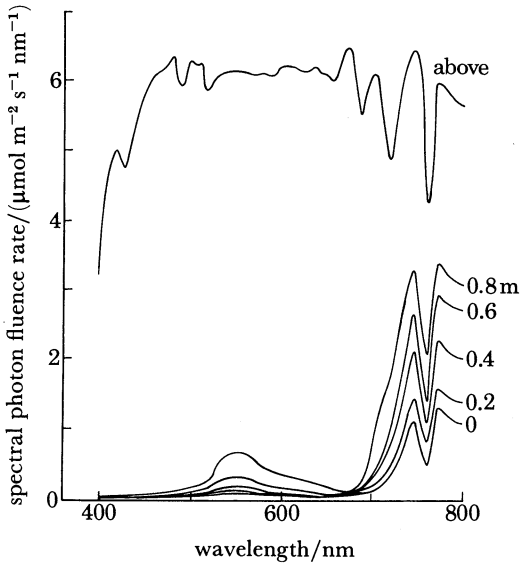

Figure 1. Spectral quality of radiation at various heights above ground level within a wheat (*Triticum aestivum* L.) canopy. The spectra were measured on 7 June 1973 with a crop height of 0.90–0.95 m under a clear sky (0/8 to 2/8 cloud cover, solar disc not obscured). (After Holmes & Smith (1977*b*).)

The alternative possibility for detecting light quality is for the plant to use a photochromic pigment, i.e. a single pigment existing in two forms that are interconvertible by light. Although photoreversible pigments which absorb in the blue–ultraviolet wavebands have been reported in fungi (Kumagai 1982; Löser & Schäfer 1980) and animals (Hamdorf *et al.* 1972), phytochrome is the only known photoreversible pigment in higher plants. There now exists extensive circumstantial evidence that the photochromic properties of phytochrome are used to detect the presence and extent of shading by other plants. The mechanism appears to depend on the absorption maxima of P_r and P_{fr} being centred around 660 and 730 nm respectively and the fact that the relative quantities of radiation in these wavebands varies under natural conditions.

(*a*) *Phytochrome action in light-grown plants*

The highest stem-elongation rates in dicotyledonous plants occur in the absence of light when either no P_{fr} has been formed (i.e. etiolation) or when the P_{fr} formed in light-grown tissue has

been removed. Phytochrome action is therefore considered to be an inhibitory effect on stem extension because any light treatment that produces P_{fr} reduces the growth rate.

Most evidence indicates that P_{fr} is the active form of phytochrome in light-grown plants. This conclusion is based on arguments similar to those used by Hendricks *et al.* (1956) for 'bulk' and 'active' phytochrome (Jabben & Holmes 1983). Although the belief that P_{fr} is the active form of phytochrome has been questioned (Smith 1981*b*; see also Smith, this volume), it will be necessary to find alternative explanations for the similarity between the action spectrum for inhibition of hypocotyl extension growth and the action spectrum for photoconversion of P_r to P_{fr} before alternatives can be considered.

Physiological experiments suggest that the P_{fr} formed in light-grown plants is very stable compared with the P_{fr} of dark-grown seedlings. An early indication of this was seen in the continued effectiveness of P_{fr} in the dark in inhibiting internode growth in *Phaseolus vulgaris* (Downs *et al.* 1957); removal of P_{fr} with far-red light during the dark period also removed the inhibition of growth that had persisted for up to 16 h after the end of the photoperiod. Similar indications of the continued effectiveness of the light received in the previous photoperiod have been monitored in the subsequent dark period by using linear displacement transducers (Lecharny & Jacques 1980; Morgan, Child & Smith, unpublished).

In *Sinapis alba*, the inhibitory effect of continuous red light on hypocotyl growth can be replaced by 5 min red light pulses interspersed by 55 min periods of darkness (Schäfer *et al.* 1981), indicating that there is no significant loss of P_{fr} over a 55 min period. However, it is clear that this P_{fr} is not completely stable because a 30 min irradiation (which gives complete photoconversion) followed by 23.5 h darkness is not as effective as 24 h continuous irradiation.

In contrast to the situation in dark-grown seedlings, phytochrome-mediated responses in light-grown dicotyledonous plants appear to be explainable on the basis of spectrophotometrically detectable phytochrome (Jabben & Holmes 1983). Phytochrome in light-grown plants is relatively stable and typically has a half-life of several hours (Butler *et al.* 1963; Koukkari & Hillman 1967; Clarkson & Hillman 1968; Wetherell 1969; Jabben & Deitzer 1978; Jabben 1980; Jabben *et al.* 1980; Atkinson *et al.* 1980; Heim *et al.* 1981; Kilsby & Johnson 1982; Rombach *et al.* 1982). Stable phytochrome derives from the apparently inactive labile pool that represents the 'bulk' phytochrome of dark-grown plants; the 'bulk' fraction is rapidly destroyed after photoconversion into P_{fr}. The phytochrome measured in plants growing in white light should not be considered to be stable unless it is demonstrated that rapid destruction does not occur after transferring the tissue to a light source that produces maximum conversion of P_r to P_{fr}. In other words, white light sources containing a high proportion of far-red light may gradually allow accumulation of labile phytochrome because the fraction held as P_r is protected from destruction.

(*b*) *Phytochrome photoequilibrium*

The variations in the relative proportions of red to far-red radiation that occur in Nature are known to cause changes in phytochrome photoequilibria *in vivo*. When etiolated plant tissue is placed on ice (to reduce the effects of phytochrome dark reactions) and exposed to a range of natural and artificial broadband spectra (Holmes & Smith 1977*a*, *b*), a hyperbolic relation is observed between the red:far-red ratio of the incident radiation and the photoequilibrium established in the tissue (Smith & Holmes 1977) (figure 2). The most striking aspect of this relation is that phytochrome exhibits greatest sensitivity to the range of changes in red:far-red that are found in natural terrestrial environments. Terrestrial plants live under situations in

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

which the red:far-red ratio varies between approximately 1.15 in full daylight and around 0.05 in dense vegetation shade (Holmes & Smith 1977*b*). It is precisely within this range that phytochrome is most sensitive to changes in light quality and would therefore act as an excellent perceiver of shade light quality.

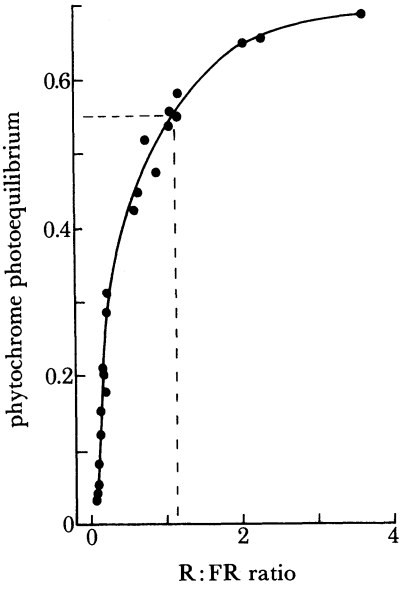

FIGURE 2. The relation between phytochrome photoequilibrium and red:far-red ratio of the incident polychromatic light. The broken line indicates photoequilibrium (*ca.* 0.54) established in natural daylight (red:far-red = *ca.* 1.15). Photoequilibria were measured in etiolated *Phaseolus vulgaris* hypocotyl hooks under both natural and artificial light sources. Both actinic irradiation and measurement of photoequilibria were done at 0 °C to reduce the effects of phytochrome dark (thermal) reactions. The figure demonstrates that phytochrome is most sensitive to changes in red:far-red ratio within the range found under natural terrestrial conditions. (After Smith & Holmes (1977).)

Two factors which might lead to deviations from the relation shown in figure 2 are the high fluence rates found in Nature and our lack of knowledge about the molecular environment of phytochrome. At high fluence rates, phytochrome exists for short periods as weakly absorbing intermediates between the P_r and P_{fr} forms (Kendrick & Spruit 1972), and under prolonged irradiation conditions the production of P_{fr} from P_r may become limited owing to the accumulation of the rate-limiting intermediate meta-Rb (Kendrick & Spruit 1976; Pratt *et al.* 1982; Spruit 1982). The molecular environment affects photoequilibrium (Mumford & Jenner 1971) and it is known that reaction rates *in vivo* are affected by both anaerobiosis (Kendrick & Spruit 1973) and by dehydration (Kendrick & Spruit 1974). However, in contrast to the situation in dark-grown tissue, most evidence points to the conclusion that phytochrome dark (i.e. non-photochemical) reactions play a relatively minor role in the establishment of photoequilibrium in light-grown plants at natural radiation levels (Jabben & Holmes 1982).

The average photoequilibrium established in green plants is lower than that in etiolated tissue because of the screening effect of chlorophyll (Holmes & Fukshansky 1979). Natural daylight, for example, establishes a photostationary state of about 0.54 in etiolated tissue whereas the average photostationary state in a green *Phaseolus* leaf illuminated on both surfaces is approximately 0.43. It should be noted, however, that although the photostationary state is

lower in green tissue, the systematic relation between the red:far-red ratio of the incident light and phytochrome photoequilibrium is not affected (Morgan & Smith 1978*a*).

(*c*) *Photoequilibrium and growth response*

Red/far-red reversible control of stem elongation in light-grown plants was first demonstrated by Downs *et al.* (1957). They showed that 5 min far-red light given at the end of an 8 h photoperiod produced longer internodes in *Phaseolus vulgaris* than plants that had received 5 min far-red immediately followed by 5 min red light. These observations were important in that they not only demonstrated the effects of these spectral regions but also showed that the response is photoreversible. Similar responses have since been recorded in several other species (see, for example, Kasperbauer 1971; Lecharny & Jacques 1974; Vince-Prue 1973, 1977; Holmes & Smith 1977*c*).

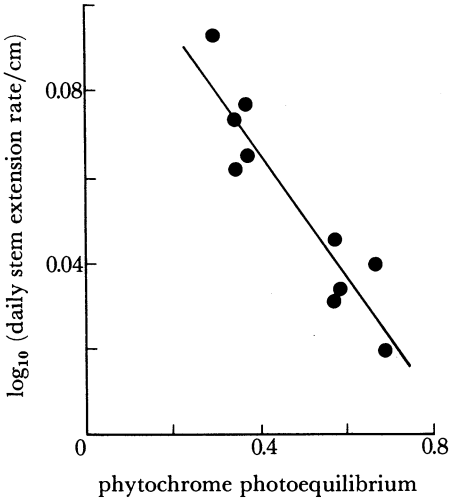

FIGURE 3. Relation between logarithmic stem extension rate in *Chenopodium album* and phytochrome photoequilibrium. Photoequilibria were derived from the curve in figure 2. The p.a.r. in all treatments was 100 μmol m^{-2} s^{-1}. The figure demonstrates the systematic increase in elongation growth rate with decreasing P_{fr}/P_{tot} ratio. (After Morgan & Smith (1976).)

It became clear from observations (Holmes & Smith 1975), in which both the quantity and quality of light in the 400–700 nm waveband was held constant, that supplementary far-red light alone causes increased stem elongation rate. These experiments indicated that the decrease in red:far-red ratio below vegetation shade, which causes a decrease in the P_{fr}/P_{tot} ratio (Holmes & Smith 1975; Smith & Holmes 1977), causes a concomitant increase in elongation growth rate (Holmes & Smith 1975, 1977*c*). A more detailed analysis of this response was made by Morgan & Smith (1976), who used a range of photoequilibria resembling those existing in natural conditions. They observed an inverse linear relation between stem growth rate and the P_{fr}/P_{tot} ratio estimated to have been established during the daily photoperiod (figure 3). These results provide circumstantial evidence that phytochrome not only detects vegetation shade, but also measures the degree of shading.

The correlation between stem extention rate and estimated photoequilibrium depicted in figure 3 extends over the range $P_{fr}/P_{tot} = 0.71$–0.26 (Morgan & Smith 1978*a*). Technical restrictions made it impossible to produce lower photoequilibria at the background fluence rate

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

of 100 μmol m^{-2} s^{-1} in the 400–700 nm waveband. Light-grown *Chenopodium rubrum* seedlings show a similar, but not identical, systematic relation between hypocotyl elongation rate and photoequilibrium over the range $P_{fr}/P_{tot} = 0.75$–0.17 (Ritter *et al.* 1981). Studies with light-grown *Sinapis alba* seedlings under monochromatic light show that this relation can hold down to P_{fr}/P_{tot} ratios as low as 0.03 (Holmes *et al.* 1982). Control experiments with chlorophyll-free plants indicated that photosynthesis was not involved in the response. A point of inflexion was observed at P_{fr}/P_{tot} lower than 0.03, where further reductions in P_{fr}/P_{tot} cause proportionally greater increases in hypocotyl elongation rate. It is noteworthy that no natural conditions are known where P_{fr}/P_{tot} ratios of less than 0.03 exist. It should also be pointed out that the growth responses of hypocotyls may not be the same as those of the growing internodes of mature plants.

The experiments described above were made at fluence rates that are known to establish photoequilibria within minutes under both the polychromatic (Holmes 1975; Smith & Holmes 1977) and monochromatic irradiation (Beggs *et al.* 1981; Jabben *et al.* 1982) conditions. At lower fluence rates, biphasic relationships between elongation response and predicted photoequilibrium are observed. Both Satter & Wetherell (1968), studying *Sinningia*, and Vince-Prue (1973, 1977), studying *Fuchsia*, found maximal elongation rates under light sources that, on the basis of photochemical reactions alone, would have produced P_{fr}/P_{tot} values in the approximate range 0.30–0.40. A similar response was found in hypocotyls of light-grown *Chenopodium rubrum*, with the highest elongation growth rate at a photostationary state of approximately 0.35 (Holmes & Wagner 1981). However, when the experiments were repeated with a tenfold increase in fluence rate, the relation between elongation rate and photostationary state became approximately linear for the entire range ($P_{fr}/P_{tot} = 0.14$ to 0.69) studied. Holmes & Wagner concluded that the biphasic response was due to the relatively high effectiveness of phytochrome dark reactions at low fluence rates (Heim & Schäfer 1982). This explanation may also account for the biphasic responses observed with *Fuchsia* (Vince-Prue 1973, 1978) and *Sinningia* (Satter & Wetherell 1968), although species differences and slight effects of photosynthesis cannot be ruled out.

Light-grown plants appear to modulate their elongation rate in immediate response to changes in light quality. In *Chenopodium album*, stem extension rate is increased within 7 min of adding monochromatic far-red light to the background white fluorescent light (Morgan & Smith 1978*b*). These rapid responses to radiation also occur after prolonged periods of darkness; *Vigna sinensis* L. responds within 30 min to monochromatic red light (Lecharny & Jacques 1980).

(*d*) *Role of the light and dark periods*

Although it is known that phytochrome controls elongation growth during the dark period (Downs *et al.* 1957) and during the light period (Holmes & Smith 1975), the relative effectiveness of these two periods is unknown. As outlined in the previous section, the qualitative responses to P_{fr} are the same in continuous light as in darkness after actinic irradiation. The responses differ in that some inhibitory effects of light are lost during long dark periods. In *Sinapis alba*, some inhibitory effect of light is lost during a 23.5 h dark period (table 1). If the dark period is reduced to 55 min, the inhibitory effect of 5 min red light pulses is as great as continuous red light. If the red light pulses are followed immediately by far-red light pulses, the response is approximately the same as with far-red light pulses alone, thereby indicating phytochrome involvement and a direct parallel with the characteristics of end-of-day responses.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Persistence of the effect of the previous light régime in darkness is also found in *Sinapis alba* plants 7 days old (table 1) and 2 weeks old (Child, Morgan & Smith, unpublished results). Similar observations have been reported by Lecharny & Jacques (1980).

In *Chenopodium rubrum*, a time-course study with both chlorophyll-free and normal green seedlings showed that a dark period of more than 12 h is required before a decrease in the inhibitory effectiveness of previous red light is observed (Holmes, Boden & Wagner, unpublished results). The response was red–far-red reversible, thereby implying phytochrome control.

TABLE 1. COMPARISON OF THE EFFECTIVENESS OF CONTINUOUS AND PULSED MONOCHROMATIC RED LIGHT (λ_{max} = 654 nm) ON INHIBITION OF HYPOCOTYL GROWTH IN LIGHT-GROWN *SINAPIS ALBA* L.

(Conditions: red light, 160 μmol m^{-2} s^{-1} for 5 min pulses, 3.3 μmol m^{-2} s^{-1} for 30 min pulse and 13 μmol m^{-2} s^{-1} for continuous light; far-red light (Schott RG9+KG1) 25 μmol m^{-2} s^{-1} below 800 nm. (After Beggs *et al.* 1981; Schäfer *et al.* 1981).)

(*a*) *Seedlings* 54 h *old*

treatment	percentage inhibition
continuous red	66.8
24 × (5 min red + 55 min darkness)	66.9
24 × (5 min red + 5 min far-red + 50 min darkness)	5.9
24 × (5 min far-red + 55 min darkness)	10.2
30 min red + 23.5 h darkness	39.0

(*b*) *Plants* 7 *days old*

treatment	daily increase in hypocotyl length/mm
continuous red	1.4
24 × (5 min red + 55 min darkness)	1.1
24 × (5 min red + 5 min far-red + 50 min darkness)	3.6
24 × (5 min far-red + 55 min darkness)	3.5

Clearly, therefore, phytochrome can maintain elongation rate in darkness for several hours under otherwise constant conditions. In Nature, however, the relative roles of light and darkness appear to be strongly influenced by other factors. Using 16 h photoperiods, Morgan & Smith (1978*a*) found that 80% of the increased growth rate in *Chenopodium album* internodes caused by supplementary far-red light was produced by the daytime irradiation. It may be significant that the 20% attributable to the night period was produced over a shorter (8 h) period and at temperatures 5 °C lower than those used during the day.

(*e*) *Ecological variability*

The many changes in ontogeny elicited by simulated canopy light confer obvious adaptive advantages to species competing with others for light. For a ruderal plant such as *Chenopodium album*, the ability to redirect development toward vertical growth at the expense of lateral growth would enable the plant to intercept more light for photosynthesis. In many shaded habitats, such as within the understory vegetation of woods and forests, increased stem extension growth would be futile. There is evidence that species adapted to shaded environments respond differently from those that grow in open habitats.

Fitter & Ashmore (1974) showed that *Veronica persica* Poir., an annual weed of disturbed

ground, radically altered its growth pattern in response to a simulated woodland light environment. In contrast to this response, *Veronica montana* L., a woodland perennial showed negligible change in its growth pattern under the same conditions.

In another study, Frankland & Letendre (1978) showed that the effects of supplementary far-red light on the growth pattern of the shade-tolerant plant *Circaea lutetiana* were not as marked as had been reported for plants adapted to open habitats. Morgan & Smith (1979) studied a wide range of species and concluded that the extent to which extension rate was modified by the prevailing red:far-red ratio was dependent on the natural habitat of the species (figure 4). Shade avoiders show marked increases in stem extension rate as the P_{fr}/P_{tot} level is decreased, whereas shade tolerators show little or no response to changes in photoequilibrium. This apparently systematic relation between natural species habitat and response to shade light quality would be of obvious value for growth and survival.

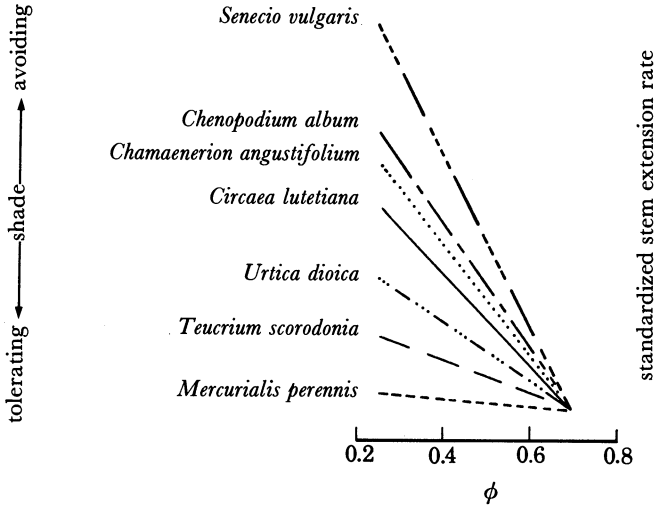

FIGURE 4. The relation between stem extension rate and phytochrome photoequilibrium. Photoequilibria were derived from the curve in figure 2. Stem extension data, expressed as logarithmic extension rate (i.e. $\log_{10}$ (growth per day)) were normalized to the value obtained at $P_{fr}/P_{tot} = 0.70$. The figure demonstrates the way in which the response to changes in red:far-red varies with the natural habitat of the species studied; the slopes of the lines for shade-avoiding species are very steep whereas those for the shade-tolerant species are very shallow. (After Morgan & Smith (1979) and Smith (1982).)

4. PERCEPTION OF LIGHT QUANTITY

Large gradients in the quantity of blue and red light exist within plant canopies. There are many examples in the literature of changes in growth and metabolism that are caused by variations in the quantity rather than the quality of light. Information is available on the effects of photon fluence rate variations at levels equivalent to sunlight (Warrington *et al.* 1978) down to the very low levels normally associated with vegetation shade (see, for example, Blackman & Wilson 1951*a*, *b*; Grime & Jeffrey 1965; Grime 1966).

Wavelength-sensitivity to changes in light quantity varies between species. In all species of light-grown plants studied in detail, it is changes in the quantity of either the blue, or the red, or both the blue and red wavebands that elicit the largest physiological responses. Two interesting examples with contrasting wavelength sensitivities are *Chenopodium rubrum* and *Sinapis*

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

alba, both of which are successful competitors for light and both of which are very sensitive to changes in fluence rate. When studied at the stage of hypocotyl extension, *S. alba* is sensitive to the quantity of red light but shows negligible response to blue, whereas *C. rubrum* is highly sensitive to the quantity of blue light but is only slightly affected by changes in the quantity of red light. The different methods by which these two species perceive changes in the quantity of light will be analysed in the following sections.

(*a*) *The blue waveband*

The relative quantities of blue and red radiation in daylight are approximately the same at solar elevations higher than about 10° (see, for example, Holmes & Smith 1977*a*), and vegetation canopies attenuate blue and red light in approximately equal proportions (see, for example, Holmes & Smith 1977*b*). As red light is 50–100 times more effective than the blue waveband in photoconverting phytochrome (Butler *et al.* 1964; Pratt & Briggs 1966; Jabben *et al.* 1982), blue light will have only minor direct effects on phytochrome under natural conditions. However, there is substantial evidence for at least one specific blue-light-absorbing photoreceptor sensitive to changes in the quantity of light.

Various arguments have been put forward to support the concept that blue light responses can be explained on the basis of phytochrome action (Hartmann 1966; Schäfer 1975; Roth-Bejerano 1980). Most evidence, however, implies the existence of a separate blue-light-absorbing photoreceptor (b.a.p.) that operates in both light-grown and dark-grown plants.

Several of the experiments that have provided evidence of a specific b.a.p. have also indicated important factors that have to be considered when determining the contribution a b.a.p. may make towards the perception of shade. The first factor is that the relative effectiveness of blue and red light in controlling developmental growth varies between species (Vince 1956; Sale & Vince 1959). Meijer (1958, 1959) compared the effectiveness of equal fluence rates of monochromatic blue, red and far-red light on internode elongation in a variety of mature green plants. In two species, elongation rate was faster in blue than red; in two other species, elongation rate was slower in blue than red; in the fifth species studied, no significant difference was detected in elongation rate. He explained this difference between species on the basis of different fluence-rate dependences of two photoreceptors. Species differences are also common in light-grown seedlings. *Lactuca sativa*, *Cucumis sativa* and *Chenopodium rubrum*, for example, are relatively sensitive to blue light (Evans *et al.* 1965; Black & Shuttleworth 1974; Ritter *et al.* 1981) whereas light-grown *Sinapis alba* show a negligible response to blue light (Wildermann *et al.* 1978).

Internode elongation rate in *Vigna sinensis* is regulated by both blue and red light (Lecharny & Jacques 1980). Using a linear displacement transducer, they found a rapid response to both blue and red light. Compared with plants held in darkness, the cumulative effect of a 10 h irradiation treatment with blue light was promotive whereas the cumulative effect of red light was slightly inhibitory. However, when the blue and red light were given simultaneously, there was a marked inhibition of growth rate relative to the rate in darkness. Their results indicated a synergistic action of the blue and red wavebands. Synergistic effects of blue and red light have also been reported for malate formation in *Vicia faba* guard cells (Ogawa *et al.* 1978) and between blue and yellow light for control of stem growth in *Pisum sativum* (Elliott 1979).

It is clear from observations such as these that whereas studies with monochromatic light

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

can increase our understanding of the individual photoreceptors that may be involved in the perception of shade, account must be made of possible interactions between photoreceptors under polychromatic irradiation conditions similar to those found in the natural environment.

As described above, sensitivity to light varies between species and organs and is also modified by age, stage and development, previous lighting conditions and, in some instances, the number of activated photoreceptors. In comparing the relative effectiveness of light quantity and quality in modifying development, it is necessary to reduce the number of variable factors for simplicity. To do this, the hypocotyl growth response to specific parameters of simulated shade light is compared in *Sinapis alba* L. and *Chenopodium rubrum* L.

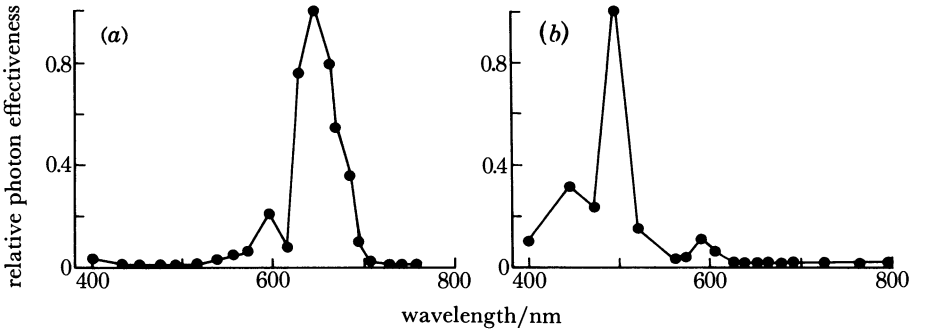

FIGURE 5. Action spectra for inhibition of hypocotyl elongation growth in (*a*) light-grown *Sinapis alba* seedlings and (*b*) light-grown *Chenopodium rubrum* seedlings. The spectra were divided from photon fluence rate curves (percentage inhibition against logarithmic photon fluence rate) for 50 % inhibition (*S. alba*) and 40 % inhibition (*C. rubrum*). The data are normalized to 1.0 at the wavelength of maximum inhibition. The action spectra illustrate differences between species in sensitivity to the blue and red wavebands. (Redrawn after Beggs *et al.* (1980) for *S. alba* and Holmes & Wagner (1982) for *C. rubrum*.)

S. alba, at the hypocotyl growth phase, responds to changes in both the quality and quantity of light in the red and far-red wavebands but shows negligible response to blue light (Figure 5), even when given as a supplement to red. In other words, the response in *Sinapis* allows a study to be made of the role played by phytochrome in perceiving shade without the additional complications produced by interacting effects of the b.a.p. *C. rubrum* is also sensitive to changes in light quality in red and far-red wavebands, but differs from *S. alba* in that it shows a relatively small response to the quantity of red or far-red light and a high sensitivity to the quantity of blue radiation (figure 5).

To determine the role played by the b.a.p. in the perception of shade it is necessary to reduce the number of variables in the actinic light. The known variables to which the plant may respond are phytochrome photoequilibrium (or concentration of P_{fr}), phytochrome cycling rate, the amount of p.a.r., the quantity of radiation in the red–far-red waveband and the quantity of radiation in the blue waveband.

The contributions of phytochrome photostationary state and cycling rate can be calculated, thereby making it possible to account for, or exclude, the contribution of these factors under either monochromatic or polychromatic radiation (Fukshansky *et al.* 1981). As a photochromic pigment, phytochrome can measure only two factors in the actinic radiations: these are the photons absorbed by P_r and the photons absorbed by P_{fr}. The product of these reactions depends on the photon fluence rate of the radiation, the quantum yield of the photoreactions

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

and the extinction coefficients of P_r and P_{fr}. The photochemical reactions of phytochrome are described by the rate constants k_1 and k_2, i.e.

$$P_r \underset{k_2}{\overset{k_1}{\rightleftharpoons}} P_{fr},$$

where

$$k_1 = N_\lambda \epsilon_{r,\lambda} \phi_{r,\lambda}$$

and

$$k_2 = N_\lambda \epsilon_{fr,\lambda} \phi_{fr,\lambda}.$$

$\epsilon_{r,\lambda}$ and $\epsilon_{fr,\lambda}$ are the extinction coefficients of P_r and P_{fr} at wavelength λ; $\phi_{r,\lambda}$ and $\phi_{fr,\lambda}$ are the quantum yields of P_r and P_{fr} phototransformation at wavelength λ; N_λ is the photon fluence rate at wavelength λ.

Under polychromatic light, the rate constants k_1 and k_2 become

$$k_1 = \int_{\lambda_1}^{\lambda_2} N_{s,\lambda} \epsilon_{r,\lambda} \phi_{r,\lambda} d\lambda$$

and

$$k_2 = \int_{\lambda_1}^{\lambda_2} N_{s,\lambda} \epsilon_{fr,\lambda} \phi_{fr,\lambda} d\lambda,$$

where $N_{s\lambda}$ is the spectral photon fluence rate.

TABLE 2. SUMMARY OF THE ACTINIC PROPERTIES OF DAYLIGHT AND CANOPY LIGHT SPECTRA, AND THEIR CALCULATED EFFECTS ON PHYTOCHROME PHOTOTRANSFORMATION KINETICS (AFTER HOLMES *ET AL.* 1982)

parameter	daylight	canopy light
k_1/min^{-1}	132.1	1.36
k_2/min^{-1}	83.6	3.75
$(k_1+k_2)/\text{min}^{-1}$	215.7	5.11
$k_1/(k_1+k_2)$	0.61	0.27
red:far-red ratio†	1.14	0.10
equivalent monochromatic wavelength/nm	687	701
equivalent monochromatic photon fluence rate/(μmol m^{-2} s^{-1})	452	17.7

† 10 nm bandwidth centred at 660 and 730 nm.

It should be noted that the photostationary state (i.e. $k_1/(k_1+k_2) = \phi$) is not the same as the P_{fr}/P_{tot} ratio established *in vivo* as a result of combined phytochrome light and dark reactions. Phytochrome dark reactions (i.e. synthesis of P_r and reversion and destruction of P_{fr}) can play a major role in determining the P_{fr}/P_{tot} ratio in dark grown plants at normal physiological temperatures (Heim & Schäfer 1981). In light-grown plants, the physiologically effective phytochrome appears to be relatively stable and phytochrome dark reactions only have a significant effect at very low fluence rates (Jabben & Holmes 1983).

The actinic properties of representative natural daylight and vegetation shade light (which are similar to the spectra in figure 1) are given in table 2. The effect that these changes in spectral quality and quantity have on hypocotyl elongation rate in *C. rubrum* can be derived from studies with artificial light sources that simulate the natural spectra by adding various amounts of far-red light to background white light (Ritter *et al.* 1981).

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

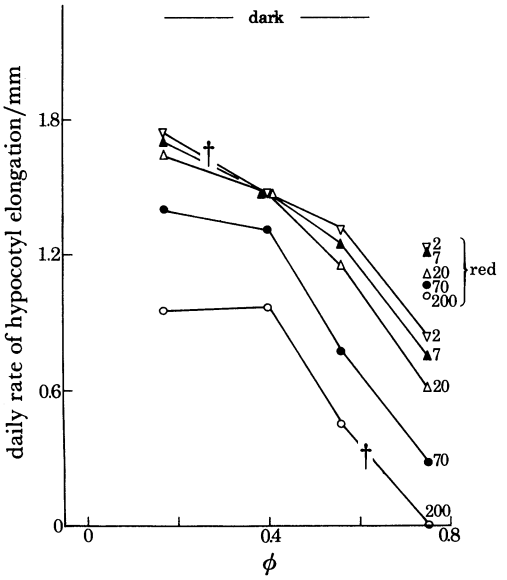

FIGURE 6. The effect of phytochrome photostationary state (ϕ) at a range of phytochrome cycling rates on hypocotyl elongation rate in chlorophyll-free *Chenopodium rubrum* seedlings. The seedlings had been in continuous white light since sowing. The reductions in ϕ were achieved by adding different amounts of far-red light to the background fluorescent white light source. The figure demonstrates that reducing the quantity (cycling rate) of monochromatic red light (red 2:2 min^{-1}, etc.) has a relatively small effect on elongation growth rate compared with reductions in the quantity of polychromatic radiation, irrespective of ϕ (2, 7, etc.: cycling rate of 2 min^{-1}, 7 min^{-1}, etc.). Reductions in ϕ also caused marked increases in growth rate. The combined effects of the reduction in the quantity of blue light, reduction in phytochrome cycling rate, and reduction in ϕ – all of which are characteristic of vegetation shade light compared with natural daylight (from table 2) – result in a large increase in elongation rate (†). The involvement of photosynthesis has been excluded by using chlorophyll-free seedlings. (After Ritter *et al.* (1981).)

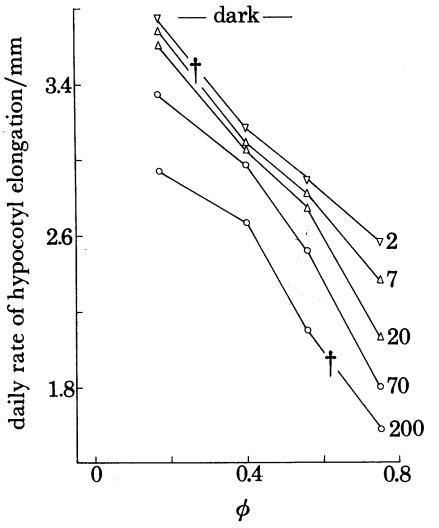

FIGURE 7. The effect of phytochrome photostationary state (ϕ) at a range of phytochrome cycling rates on hypocotyl elongation rate in green *Chenopodium rubrum* seedlings. The seedlings had been in continuous white light since sowing. The reductions in ϕ were achieved by adding different amounts of far-red light to the background white light source. Reducing the quantity of monochromatic red light does not have a consistent effect on hypocotyl growth rate, whereas reducing the quantity of polychromatic radiation causes a marked increase in elongation rate at all ϕ values studied (2, 7, etc.: cycling rate of 2 min^{-1}, 7 min^{-1}, etc.). Reductions in ϕ also cause marked increases in growth rate. The combined effects of the reduction in the quantity of blue light, reduction in phytochrome cycling rate, and reduction in ϕ characteristic of vegetation shade light compared with natural daylight (from table 2) result in a large increase in elongation rate (†). (After Ritter *et al.* (1981).)

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

The effect of activating the b.a.p. on the response of hypocotyl elongation rate to changes in ϕ is seen in figure 6. The data are plotted for five equal phytochrome cycling rates and the effects of photosynthesis are eliminated by using chlorophyll-free seedlings. Two important points are evident. First, whereas reducing the quantity (indicated by cycling rate) of monochromatic red light (or far-red, not shown) causes only a small increase in elongation rate, a reduction in the quantity of polychromatic radiation causes a relatively large increase in growth rate at all ϕ tested. Second, reducing ϕ at a constant cycling rate also causes an increase in elongation rate; this increase is not caused by the slightly higher level of blue and red light used to maintain constant cycling rate because the same response is observed when blue and red are constant (Holmes & Wagner 1981, 1982; Ritter *et al.* 1981). Qualitatively similar responses were found in green seedlings (figure 7).

The roles played by the b.a.p. and by phytochrome in the perception of shade by *C. rubrum* can be interpolated from the curves in figures 6 and 7 by comparing the actinic properties of the experimental light treatments with the actinic properties of daylight and canopy shade light (table 2). It can be seen that the elongation rate of green seedlings (figure 7) in light equivalent to canopy shade († at $\phi = 0.27$ in figure 7) is increased relative to the rate in daylight († at $\phi = 0.61$ in figure 7) for two reasons. First, there is an increase due to the reduction in ϕ. Second, there is an increase due to the reduction in fluence rate (expressed as reduced cycling rate) and this increase is only significant in the presence of blue light. In other words, the reduced photon fluence rate of blue light below vegetation shade relative to daylight is perceived by a b.a.p. and this photoreceptor plays a major role in modulating elongation growth rate in *C. rubrum*.

(*b*) *The red waveband*

(i) *Phytochrome as photoreceptor*

There is no unequivocal evidence for phytochrome-modulated fluence-rate dependence of elongation growth (i.e. internode) in mature green plants. Although a fluence-rate dependence does exist in green plants in so far as red light affects elongation rate relative to darkness, the relation between the effects of photosynthesis and the effects of phytochrome is unknown.

In light-grown seedlings, phytochrome-modulated hypocotyl growth responses exhibit fluence-rate dependence and the effects caused by photosynthesis and by phytochrome can be separated. The extent of the fluence-rate dependence and the range over which it occurs differs greatly between species. Hypocotyl elongation rate in light-grown *Sinapis alba* seedlings is strongly dependent on the fluence rate in the red waveband (Wildermann *et al.* 1978; Beggs *et al.* 1980). The response cannot be attributed to photosynthesis because chlorophyll-free plants also exhibit a fluence-rate dependence.

There is correlative evidence in dark-grown *S. alba* seedlings that the fluence-rate dependent component for inhibition of hypocotyl growth by monochromatic red light represents, at least in part, the phytochrome photoconversion processes competing against the phytochrome dark reactions (Heim & Schäfer 1982), and a similar mechanism has been proposed to operate in light-grown plants (Jabben & Holmes 1983). However, at least one factor other than photoconversion must be involved because some fluence-rate dependent response is observed at fluence rates higher than those required for complete photoconversion of P_r to P_{fr} (Beggs *et al.* 1980; Heim & Schäfer 1982).

Cycling of phytochrome has been proposed as a possible mechanism to explain phytochrome perception of light quantity (see, for example, Jose & Vince-Prue 1978; Johnson & Tasker

1979), although no empirical evidence for this hypothesis has yet been found. In light-grown *S. alba* seedlings, there is no correlation between cycling rate and response (Holmes *et al.* 1982). Morgan *et al.* (1980) used a specific test for the involvement of cycling in regulation of stem elongation in mature *S. alba* plants. They noted that stem extension rate was increased by various wavelengths of far-red light added to a background of white light and that the response was enhanced by increasing the far-red fluence rate. The involvement of cycling *per se* was excluded because addition of monochromatic red light to the white light plus far-red resulted in a decrease, rather than a further increase, in extension rate. Morgan *et al.* (1981) provided further evidence that cycling is not involved in stem extension in mature *S. alba* and that the response to supplementary far-red light is not fluence-rate dependent.

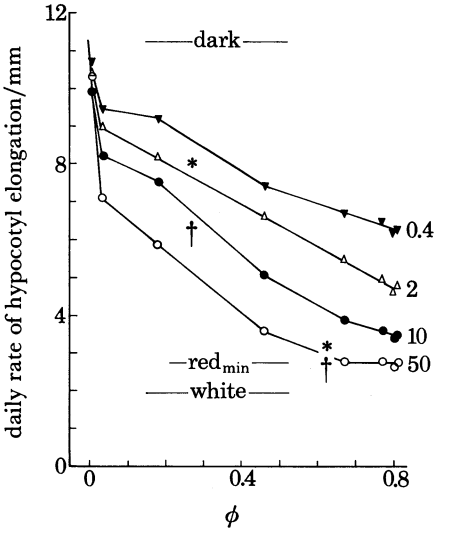

FIGURE 8. The effects of phytochrome photostationary state and photon fluence rate on hypocotyl elongation rate in light-grown *Sinapis alba*: ○, 50 μmol m^{-2} s^{-1}; ●, 10 μmol m^{-2} s^{-1}; △, 2.0 μmol m^{-2} s^{-1}; ▼, 0.4 μmol m^{-2} s^{-1}. The line labelled red$_{min}$ indicates the slowest growth rate (i.e. maximum inhibition) that can be obtained in the red–far-red waveband; the line labelled white indicates the slowest growth rate that can be obtained with white xenon arc light (*ca.* 300 μmol m^{-2} s^{-1} in the 400–800 nm waveband); the small difference between red$_{min}$ and white shows that the b.a.p. makes only a very small contribution to the modulation of elongation growth rate. The figure demonstrates that both a reduction in the red:far-red ratio and a reduction in the quantity of light in the red–far-red waveband contribute to modulation of hypocotyl growth rate. Phytochrome-modulated growth rates are presented for full daylight and canopy light (†, see table 2) and for 10% of daylight and canopy light (*). (After Holmes *et al.* (1982).)

(ii) *Phytochrome in Sinapis alba L.*

Light-grown *S. alba* differs from light-grown *C. rubrum* in that the hypocotyl growth rate shows little response to the quantity of blue light but a marked response to changes in the quantity of red light (Wildermann *et al.* 1978; Beggs *et al.* 1980; Schafer *et al.* 1981). The role played by the quantity of light in the red waveband in the perception of shade can be determined by comparing the actinic properties for phytochrome of daylight and canopy shade light (table 2) with the response of *S. alba* hypocotyls to monochromatic red and far-red light with the equivalent actinic properties (figure 8). Clearly, all wavelengths, or combinations of wavelengths, that produce the same phytochrome photostationary state and cycling rate will be perceived as identical by the plant (Fukshansky *et al.* 1981). For example, a single wavelength that

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

produces a photostationary state of $\phi = 0.30$ is identical for the phytochrome in the plant to a combination of wavelengths (e.g. red plus far-red) that produce the same photostationary state and the same cycling state.

The same phytochrome photoequilibrium established by daylight is produced by monochromatic light at 687 nm (table 2). To obtain the same cycling rate, a photon fluence rate of 452 μmol m^{-2} s^{-1} would be required. The shade light is equivalent for the phytochrome system to monochromatic 701 nm radiation at a photon fluence rate of 17.7 μmol m^{-2} s^{-1}.

The data in figure 8 show that *S. alba* hypocotyl extension growth is controlled by both the quantity and the quality of radiation in the red and far-red wavebands. The photostationary state and cycling rate produced by daylight results in an elongation rate of 2.7 mm per day whereas light equivalent to canopy shade produces an elongation rate of 6.4 mm per day. The important point to note is that decreasing ϕ alone does not produce such a large increase in elongation rate as the combined effects of decreasing ϕ and the quantity of light absorbed by phytochrome. If the fluence rate of both the daylight and canopy light is reduced to 10%, the role played by the quantity of light is increased.

It must be emphasized that this approach deliberately excludes the small contribution that blue light makes to the response (indicated by the difference between red_{min} and white in figure 8) and indicates only the two ways by which phytochrome can perceive shade.

5. Conclusions

Evidence has been presented that supports the concept that a fundamental function of phytochrome is to perceive the natural light environment and to modify growth and development accordingly. The photochromic properties of phytochrome enable a plant to respond specifically to changes in light quality caused by surrounding vegetation. This ability to acquire information on the type of shading is of obvious adaptive and survival value as it would allow the plant to react appropriately to competition.

Apart from indicating the extent and nature of shade, phytochrome also functions in some species to detect changes in the fluence rate. Other species appear to be primarily dependent on one or more blue-light-absorbing photoreceptors to detect changes in fluence rate. The perception of changes in fluence rate is important because it enables a plant to respond to both vegetation shade and shading caused by objects that have only a small effect on spectral quality, such as stones and soil. It is noteworthy that the response to fluence rate is immediate and marked in the initial stages of elongation growth (hypocotyl) but plays a lesser role in modulating growth rate in the adult plant (internode). The response to light quality differs in that it is developed only after a few hours of receiving light and that the sensitivity to light quality persists to play the major role in controlling elongation growth in the adult plant.

References

Atkinson, Y. E., Bradbeer, J. W. & Frankland, B. 1980 In *Photoreceptors and plant development* (ed. J. deGreef), pp. 543–550. Antwerpen University Press.

Beggs, C. J., Geile, W., Holmes, M. G., Jabben, M., Jose, A. M. & Schäfer, E. 1981 High irradiance response promotion of a subsequent light induction response in *Sinapis alba* L. *Planta* **151**, 135–140.

Beggs, C. J., Holmes, M. G., Jabben, M. & Schäfer, E. 1980 Action spectra for the inhibition of hypocotyl growth by continuous irradiation in light and dark-grown *Sinapis alba* L. seedlings. *Pl. Physiol.* **66**, 615–618.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Black, M. & Shuttleworth, J. E. 1974 The role of the cotyledons in the photocontrol of hypocotyl extension in *Cucumis sativa* L. *Planta* **117**, 57–66.
Blackman, G. E. & Wilson, G. L. 1951*a* Physiological and ecological studies in the analysis of plant environment. VI. The constancy for different species of a logarithmic relationship between net assimilation rate and light intensity and its ecological significance. *Ann. Bot.* **15**, 63–94.
Blackman, G. E. & Wilson, G. L. 1951*b* Physiological and ecological studies in the analysis of plant environment. VII. An analysis of the differential effects of light intensity on the net assimilation rate, leaf-area and relative growth rate of different species. *Ann. Bot.* **15**, 373–408.
Butler, W. L., Lane, H. C. & Siegelman, H. W. 1963 Non-photochemical transformations of phytochrome *in vivo*. *Pl. Physiol.* **38**, 514–519.
Butler, W. L., Siegelman, H. W. & Miller, C. O. 1964 Denaturation of phytochrome. *Biochemistry, Wash.* **3**, 851–857.
Child, R., Morgan, D. C. & Smith, H. 1981 Control of development in *Chenopodium album* L. by shadelight. The effect of light quality (red:far-red ratio) on morphogenesis. *New Phytol.* **89**, 545–555.
Clarkson, D. T. & Hillman, W. S. 1968 Stable concentrations of phytochrome in *Pisum* under continuous illumination with red light. *Pl. Physiol.* **43**, 88–92.
Downs, R. J., Hendricks, S. B., Borthwick, H. A. 1957 Photoreversible control of elongation in Pinto beans and other plants under normal conditions of growth. *Bot. Gaz.* **118**, 199–208.
Evans, L. T., Hendricks, S. B. & Borthwick, H. A. 1965 The role of light in suppressing hypocotyl elongation in lettuce and petunia. *Planta* **64**, 201–218.
Elliott, W. M. 1979 Control of leaf and stem growth in light-grown pea seedlings by two high irradiance responses. *Pl. Physiol.* **63**, 833–836.
Federer, C. A. & Tanner, C. B. 1966 Spectral distribution of light in the forest. *Ecology* **47**, 555–560.
Fitter, A. H. & Ashmore, C. J. 1974 Response of two *Veronica* spp. to a simulated woodland light climate. *New Phytol.* **73**, 997–1001.
Frankland, B. & Letendre, R. J. 1978 Phytochrome and effects of shading on growth of woodland plants. *Photochem. Photobiol.* **27**, 223–230.
Fukshansky, L., Beggs, C. J., Holmes, M. G., Jabben, M. & Schäfer, E. 1981 *Proc. Eur. Symp. Light Mediat. Plant Dev.* no. 9, p. 14.
Grime, J. P. 1966 Shade avoidance and shade tolerance in flowering plants. In *Light as an ecological factor* (ed. R. Bainbridge, G. C. Evans & O. Rackham), pp. 187–207. Oxford: Blackwell.
Grime, J. P. 1981 Plant strategies in shade. In *Plants and the daylight spectrum* (ed. H. Smith), pp. 159–186. London: Academic Press.
Grime, J. P. & Jeffrey, D. W. 1965 Seedling establishment in vertical gradients of sunlight. *J. Ecol.* **53**, 621–642.
Hamdorf, K., Paulsen, R., Schwemer, J. & Tauber, U. 1972 Photoconversion of invertebrate visual pigments. In *Information processing in the visual systems of arthropods* (ed. P. Wehner), pp. 97–108. Berlin: Springer.
Hartmann, K. M. 1966 A general hypothesis to interpret 'high energy phenomena' of photomorphogenesis on the basis of phytochrome. *Photochem. Photobiol.* **5**, 349–366.
Heim, B., Jabben, M. & Schäfer, E. 1981 Phytochrome destruction in dark- and light-grown *Amaranthus candatus* seedlings. *Photochem. Photobiol.* **34**, 89–93.
Heim B. & Schäfer, E. 1982 Light-controlled inhibition of hypocotyl growth in *Sinapis alba* L. seedlings. Fluence rate dependence of hourly light pulses and continuous irradiation. *Planta* **154**, 150–155.
Hendricks, S. B., Borthwick, H. A. & Downs, R. J. 1956 Pigment conversion in the formative responses of plants to radiation. *Proc. natn. Acad. Sci. U.S.A.* **42**, 19–26.
Holmes, M. G. 1975 Studies on the ecological significance of phytochrome. Ph.D. thesis, University of Nottingham.
Holmes, M. G. 1981 Spectral distribution of radiation within plant canopies. In *Plants and the daylight spectrum* (ed. H. Smith), pp. 148–158. London: Academic Press.
Holmes, M. G., Beggs, C. J., Jabben, M. & Schäfer, E. 1982 Hypocotyl growth in *Sinapis alba* L.: the roles of light quality and quantity. *Pl. Cell Environ.* **5**, 45–51.
Holmes, M. G. & Fukshansky, L. 1979 Phytochrome photoequilibria in green leaves under polychromatic radiation: a theoretical approach. *Pl. Cell Environ.* **2**, 59–66.
Holmes, M. G. & McCartney, H. A. 1976 Spectral energy distribution in the natural environment and its implications for phytochrome function. In *Light and plant development* (ed. H. Smith), pp. 467–476. London: Butterworth.
Holmes, M. G. & Smith, H. 1975 The function of phytochrome in plants growing in the natural environment. *Nature, Lond.* **254**, 512–514.
Holmes, M. G. & Smith, H. 1977*a* The function of phytochrome in the natural environment. I. Characterization of daylight for studies in photomorphogenesis and photoperiodism. *Photochem. Photobiol.* **25**, 533–538.
Holmes, M. G. & Smith, H. 1977*b* The function of phytochrome in the natural environment. II. The influence of vegetation canopies on the spectral energy distribution of natural daylight. *Photochem. Photobiol.* **25**, 539–545.
Holmes, M. G. & Smith, H. 1977*c* The function of phytochrome in the natural environment. IV. Light quality and plant development. *Photochem. Photobiol.* **25**, 551–557.
Holmes, M. G. & Wagner, E. 1981 Phytochrome control of hypocotyl extension in light-grown *Chenopodium rubrum* L. *Physiologia Pl.* **53**, 233–238.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Holmes, M. G. & Wagner, E. 1982 The influence of chlorophyll on the spectral control of elongation growth in *Chenopodium rubrum* L. hypocotyls. *Pl. Cell Physiol.* **23**, 745–750.
Jabben, M. 1980 The phytochrome system in light-grown *Zea mays* L. *Planta* **149**, 91–96.
Jabben, M., Beggs, C. & Schäfer, E. 1982 Dependence of P_{fr}/P_{tot} ratios on light quality and light quantity. *Photochem. Photobiol.* **35**, 709–712.
Jabben, M. & Deitzer, G. F. 1978 Spectrophotometric phytochrome measurements in light-grown *Avena sativa* L. *Planta* **143**, 309–313.
Jabben, M., Heim, B. & Schäfer, E. 1980 The phytochrome system in light- and dark-grown dicotyledonous seedlings. in *Photoreceptors and plant development* (ed. J. A. deGreef), pp. 135–158. Antwerpen University Press.
Jabben, M. & Holmes, M. G. 1983 Phytochrome in light-grown plants. In *Encyclopedia of plant physiology*, vol. 16 (*Photomorphogenesis*) (ed. W. Shropshire, Jr & H. Mohr), ch. 27. Berlin: Springer. (In the press.)
Johnson, C. B. 1981 How does phytochrome perceive light quality? In *Plants and the daylight spectrum* (ed. H. Smith), pp. 481–497. London: Academic Press.
Johnson, C. B. & Tasker, R. 1979 A scheme to account quantatively for the action of phytochrome in etiolated and light-grown plants. *Pl. Cell Environ.* **2**, 259–265.
Jose, A. M. & Vince-Prue, D. 1978 Phytochrome action: a reappraisal. *Photochem. Photobiol.* **27**, 209–216.
Kasperbauer, M. J. 1971 Spectral distribution of light in a tobacco canopy and effects of end-of-day light quality on growth and development. *Pl. Physiol.* **47**, 775–778.
Kendrick, R. E. & Spruit, C. J. P. 1972 Light maintains high levels of phytochrome intermediates. *Nature, new Biol.* **237**, 281–282.
Kendrick, R. E. & Spruit, C. J. P. 1973 Phytochrome intermediates *in vivo*. I. Effects of temperature, light intensity, wavelength and oxygen on intermediate accumulation. *Photochem. Photobiol.* **18**, 139–144.
Kendrick, R. E. & Spruit, C. J. P. 1974 Inverse dark reversion of phytochrome: an explanation. *Planta* **120**, 265–272.
Kendrick, R. E. & Spruit, C. J. P. 1976 Intermediates in the photoconversion of phytochrome. In *Light and plant development* (ed. H. Smith), pp. 31–43. London: Butterworth.
Kilsby, C. A. H. & Johnson, C. B. 1982 The *in vivo* spectrophotometric assay of phytochrome in two mature dicotyledonous plants. *Photochem. Photobiol.* **35**, 255–260.
Koukkari, W. L. & Hillman, W. S. 1967 Effects of temperature and aeration on phytochrome transformations in *Pastinaca sativa* root tissue. *Am. J. Bot.* **59**, 1118–1122.
Kumagai, T. 1982 Blue and near ultraviolet reversible photoreaction in the induction of fungal conidiation. *Photochem. Photobiol.* **35**, 123–125.
Lecharny, A. & Jacques, R. 1974 Phytochrome et croissance des tiges; variations de l'effet de la lumière en fonction du temps et du lieu de photoperception. *Physiol. vég.* **12**, 721–738.
Lecharny, A. & Jacques, R. 1979 Phytochrome and internode elongation in *Chenopodium polyspesmum* L. The light fluence rate during the day and the end-of-day effect. *Planta* **146**, 575–577.
Lecharny, A. & Jacques, R. 1980 Light inhibition of internode elongation in green plants. A kinetic study with *Vigna sinensis* L. *Planta* **149**, 384–388.
Löser, G. & Schäfer, E. 1980 Phototropism in *Phycomyces*: a photochromic sensor pigment? In *The blue light syndrome* (ed. H. Senger), pp. 244–250. Berlin: Springer.
McLaren, D. F. & Smith, H. 1978 Phytochrome control of the growth and development of *Rumex obtusifolius* under simulated canopy light environments. *Pl. Cell Environ.* **1**, 61–67.
Meijer, G. 1958 Influence of light on the elongation of gherkin seedlings. *Acta bot. neerl.* **7**, 614–620.
Meijer, G. 1959 Spectral dependence of flowering and elongation. *Acta bot. neerl.* **8**, 189–246.
Mohr, H. 1972 *Lectures on photomorphogenesis*. (237 pages.) New York: Springer.
Mohr, H. 1980 Interaction between blue light and phytochrome in photomorphogenesis. In *The blue light syndrome* (ed. H. Senger), pp. 97–109. Berlin: Springer.
Morgan, D. C., Child, R. & Smith, H. 1981 Absence of fluence rate dependency of phytochrome modulation of stem extension in light-grown *Sinapis alba* L. *Planta* **151**, 497–498.
Morgan, D. C., O'Brien, T. & Smith, H. 1980 Rapid photomodulation of stem extension in *Sinapis alba* L. Studies on kinetics, site of perception and photoreceptor. *Planta* **150**, 95–101.
Morgan, D. C. & Smith, H. 1976 Linear relationship between phytochrome photoequilibrium and growth in plants under simulated natural radiation. *Nature, Lond.* **262**, 210–212.
Morgan, D. C. & Smith, H. 1978*a* The relationship between phytochrome photoequilibrium and development in light grown *Chenopodium album* L. *Planta* **142**, 187–193.
Morgan, D. C. & Smith, H. 1978*b* Simulated sunflecks have large, rapid effects on plant stem extension. *Nature, Lond.* **273**, 534–536.
Morgan, D. C. & Smith, H. 1979 A systematic relationship between phytochrome-controlled development and species habitat, for plants grown in simulated natural radiation. *Planta* **145**, 253–258.
Morgan, D. C. & Smith, H. 1981*a* Control of development in *Chenopodium album* L. by shadelight: the effect of light quantity (total fluence rate) and light quality (red:far-red ratio). *New Phytol.* **88**, 239–248.
Morgan, D. C. & Smith, H. 1981*b* Non-photosynthetic responses to light quality. In *Encyclopedia of plant physiology*, vol. 12a (ed. P. Nobel), pp. 109–134. Springer, Berlin.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Mumford, F. E. & Jenner, E. L. 1971 Catalysis of the phytochrome dark reaction by reducing agents. *Biochemistry, Wash.* **10**, 98–101.
Ogawa, T., Ishikawa, H., Shimada, K. & Shibata, K. 1978 Synergistic action of red and blue light and action spectrum for malate formation in guard cells of *Vicia faba* L. *Planta* **142**, 61–65.
Pratt, L. H. & Briggs, W. R. 1966 Photochemical and nonphotochemical reactions of phytochrome *in vivo*. *Pl. Physiol.* **41**, 467–474.
Pratt, L. H., Shimazaki, Y., Inoue, Y. & Furuya, M. 1982 Analysis of phototransformation intermediates in the pathway from the red-absorbing to the far-red-absorbing form of *Avena* phytochrome by a multichannel transient spectrum analyzer. *Photochem. Photobiol.* **36**, 471–477.
Ritter, A., Wagner, E. & Holmes, M. G. 1981 Light quantity and quality interactions in the control of elongation growth in light-grown *Chenopodium rubrum* L. seedlings. *Planta* **153**, 556–560.
Rombach, J., Bensink, J. & Katsura, N. 1982 Phytochrome in *Pharbitis nil* during and after de-etiolation. *Physiologia Pl.* **56**, 251–258.
Roth-Bejerano, N. 1980 Growth control by phytochrome of de-etiolated bean hypocotyls mediated by chlorophyll fluorescence. *Physiologia Pl.* **50**, 326–330.
Sale, P. J. M. & Vince, D. 1959 Effects of wavelength and time of irradiation on internode length in *Pisum sativum* and *Tropaeoleum majus*. *Nature, Lond.* **183**, 1174–1175.
Satter, R. L. & Wetherell, D. F. 1968 Photomorphogenesis in *Sinningia speciosa*, cv. Queen Victoria. II. Stem elongation: interaction of a phytochrome-controlled process and a red-requiring, energy dependent reaction. *Pl. Physiol.* **43**, 961–967.
Schäfer, E. 1975 A new approach to explain the 'high irradiance responses' of photomorphogenesis on the basis of phytochrome. *J. math. Biol.* **2**, 41–56.
Schäfer, E., Beggs, C. J., Fukshansky, L., Holmes, M. G. & Jabben, M. 1981 A comparative study of the responsivity of *Sinapis alba* L. seedlings to pulsed and continuous irradiation. *Planta* **253**, 258–261.
Schmidt, R. & Mohr, H. 1982 Evidence that a mustard seedling responds to the amount of P_{fr} and not to the P_{fr}/P_{tot} ratio. *Pl. Cell Environ.* **5**, 495–499.
Smith, H. 1975 *Phytochrome and photomorphogenesis*. London: McGraw-Hill.
Smith, H. 1981*a* Function, evolution and action of plant photosensors. In *Plants and the daylight spectrum* (ed. H. Smith), pp. 499–508. London: Academic Press.
Smith, H. 1981*b* Evidence that P_{fr} is not the active form of phytochrome in light grown maize. *Nature, Lond.* **293**, 163–165.
Smith, H. 1982 Light quality, photoperception and plant strategy. *A. Rev. Pl. Physiol.* **33**, 481–518.
Smith, H. & Holmes, M. G. 1977 The function of phytochrome in the natural environment III. Measurement and calculation of phytochrome photoequilibrium. *Photochem. Photobiol.* **25**, 547–550.
Smith, H. & Morgan, D. C. 1982 The function of phytochrome in nature. In *Encyclopedia of plant physiology*, vol. 16 (*Photomorphogenesis*) (ed. W. Shropshire, Jr & H. Mohr). Berlin: Springer. (In the press.)
Spruit, C. J. P. 1982 Phytochrome intermediates *in vivo*. IV. Kinetics of P_{fr} emergence. *Photochem. Photobiol.* **35**, 117–121.
Stoutjesdijk, P. 1972*a* Spectral transmission curves of some types of leaf canopies with a note on seed germination. *Acta bot. neerl.* **21**, 185–191.
Stoutjesdijk, P. 1972*b* A note on the spectral transmission of light by tropical rainforest. *Acta bot. neerl.* **21**, 346–350.
Tasker, R. & Smith, H. 1977 The function of phytochrome in the natural environment. V. Seasonal changes in the radiant energy quality in woodlands. *Photochem. Photobiol.* **26**, 487–491.
Thomas, B. 1981 Specific effects of blue light on plant growth and development. In *Plants and the daylight spectrum* (ed. H. Smith), pp. 443–459. London: Academic Press.
Vezina, P. E. & Boulter, D. W. K. 1966 The spectral composition of near UV and visible radiation beneath forest canopies. *Can. J. Bot.* **44**, 1264–1267.
Vince, D. 1956 Studies on the effects of light quality on the growth and development of plants. II. Formative effects in *Lycopersicon esculentum* and *Pisum sativum*. *J. hort. Sci.* **31**, 16–24.
Vince-Prue, D. 1973 Phytochrome and the natural light environment. *Anais Acad. bras. Cienc.* **45** (suppl.), 93–102.
Vince-Prue, D. 1977 Photocontrol of stem elongation in light-grown plants of *Fuchsia hybrida*. *Planta* **133**, 149–156.
Wall, J. K. & Johnson, C. B. 1981 Phytochrome action in light-grown plants: the influence of light quality and fluence rate on extension growth in *Sinapis alba* L. *Planta* **153**, 101–108.
Warrington, I. J., Edge, E. A. & Green, L. M. 1978 Plant growth under high radiant energy fluxes. *Ann. Bot.* **42**, 1305–1313.
Wetherell, D. F. 1969 Phytochrome in cultured wild carrot tissue. I. Synthesis. *Pl. Physiol.* **44**, 1734–1737.
Wildermann, A., Drumm, H., Schäfer, E. & Mohr, H. 1978 Control by light of hypocotyl growth in de-etiolated mustard seedlings. I. Phytochrome as the only photoreceptor pigment. *Planta* **141**, 211–216.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Discussion

A. M. Farmer (*Botany Department, The University, St Andrews, U.K.*). How does Dr Holmes consider that aquatic plants perceive shade? One species of aquatic plant may live in three very different conditions, as regards red and far-red light: (*a*) in deep water shade where the red/far-red ratio is very high; (*b*) in shallow water, but under the canopy of other plants so that it is experiencing green shade, with a low red/far-red ratio; (*c*) in a combination of both green shade and aquatic shade such that the red/far-red ratio is similar to that in the surface irradiance.

However, the resultant shading in each of these conditions may reduce carbon assimilation by a similar degree, and so in this respect the plants are in ecologically comparable situations.

M. G. Holmes. It is unlikely that phytochrome is responsible for perception of shade caused by water alone in deep aquatic habitats because the changes in red/far-red ratio are not over a range to which phytochrome is very sensitive. In shallow water, however, the variation in red/far-red ratio can be greater (especially where there is an overlying leafy canopy) and phytochrome is ideally suited for the perception of such spectral variations. The increased proportion of far-red light near the surface relative to deep water would be sufficient to induce a relatively large change in phytochrome photoequilibrium and thereby account for morphogenetic changes such as the development of aerial fronds. A blue-light-absorbing photoreceptor would lead to less confusion in both deep and shallow water. It would be naïve to assume that plants, both terrestrial and aquatic, possess only the signal-transducing photoreceptors that we have so far discovered.

M. R. Bartley (*Department of Botany, The University, St Andrews, U.K.*). Given that although spectral changes of light penetrating water result in substantial increases in red/far-red ratio with depth, but with little change in the predicted phytochrome photoequilibrium from shallow to deep water, would Dr Holmes consider it possible that the rate of phytochrome cycling (which would decrease dramatically with depth) might play a role in controlling photomorphogenesis underwater, as it does in the photocontrol of seed germination?

M. G. Holmes. The possibility that phytochrome cycling might play a role in controlling photomorphogenesis cannot be excluded. Unfortunately, no extensive studies have been made in aquatic plants. However, this question has been approached in terrestrial plants. With the exception of photocontrol of seed germination, which Dr Bartley mentioned, the findings so far have provided no empirical evidence for the involvement of phytochrome cycling *per se* in photomorphogenesis.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES