

## The oligodynamic action of light on the germination of turions of *Spirodela polyrrhiza* (L.) Schleiden

by

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### INTRODUCTION

Previous physiological investigations on the germination of turions of *Spirodela polyrrhiza* (Czopek 1959b) showed that light is not an indispensable trophic factor. It acts, however, as a photochemical agent stimulating their germination. The *Spirodela* turions are specially sensitive to light. In darkness they are able to germinate in a low percentage (about 35%), but a 10 minutes long illumination is sufficient to rise the germination percentage to the same level as in continuous light (about 100%). Thus light plays the part of an oligodynamic factor, i.e. a factor characterized by an disproportion between its low intensity and its effect. A similar oligodynamic action of light on germination was observed — among others — in *Lepidium* seeds (Toole *et al.* 1955), which either do not germinate in darkness at all, or if so, at a very low percentage. An 8 minutes long illumination with red light is sufficient for the *Lepidium* seeds to germinate to the same extent as in continuous light. The effect of red light could be reversed by an immediate treatment with far red radiation. The phenomenon was already observed by Borthwick *et al.* (1952) in their study on the action of red and far red light on the germination of lettuce seeds. Since then presence in plants of an reversible photochemical system sensitive to red and far red radiations has been discovered by many investigators. This photoreversible system participates in many photobiological processes such as the control of photoperiodic flowering, deetiolation, germination of seeds and spores, pigment formation in tomato fruits cuticle and others (Wassink and Stolwijk 1956). The oligodynamic action of light on the germination of turions (Czopek 1959b) permits to suppose that the same absorption system is a factor stimulating the germination of turions.

Relatively little is known about the physiology of turions of *Spirodela polyrrhiza*. There are a few papers dealing with the formation of turions, their dormancy stage (Jacobs 1947, Henssen 1954, Landolt 1957, Czopek 1959b, Hillman 1961) and their germination ability (Jacobs 1947, Czopek 1959b).

In this paper are summarized the results of an investigation carried out on two problems concerning the germination of turions of *Spirodela polyrrhiza*, viz.:

1. The action of various spectral regions of light on the germination rate of turions,
2. The reversibility of the biological photoreaction red — far red.

#### MATERIAL AND METHODS

Turions of *Spirodela polyrrhiza* provided the experimental material used in these investigations. They derived from sterile laboratory cultures (Czopek 1959a) grown on Pirson and Seidel's nutrient solution with 1% sucrose. The turions were stored in darkness for one month in a refrigerator (temperature 0—3°C); they were immersed in

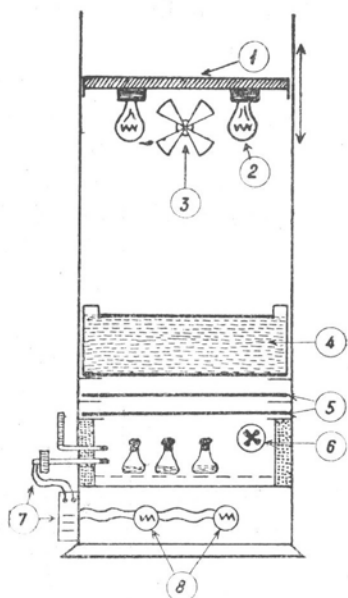


Fig. 1. Scheme of the thermostat used in experiments on the action of chromatic light on turion germination. 1 — movable plate; 2 — light source; 3 — ventilator; 4 — glass container with a liquid filter; 5 — coloured glass filters; 6 — ventilator; 7 — contact thermometer with a mercury relay; 8 — heating arrangement (bulbs)

sterile water that was periodically changed. They were subsequently transferred into Erlenmeyer flasks containing 25 ml anorganic Pirson and Seidel's nutrient solution (Kandeler 1955). 150 turions were used for each experimental series, each flask containing 15 turions. All the experiments were performed in triplicate.

In some series the turions were allowed to germinate in a light thermostat at  $28^{\circ}\text{C}$  ( $\pm 1^{\circ}\text{C}$ ) in white light emitted by 6 fluorescent tubes, 25 W. The light intensity was  $10\ 660\text{ ergs/cm}^2/\text{sec}$ . In other experiments the turions germinated in thermostats at the same temperature either exposed to chromatic light (the details are given below) or in complete darkness. The number of germinated turions was determined every day from the beginning till the end of the germination process. Turions which formed 1—3 roots and a daughter frond (1—1.5 mm long) and were able to emerge to the surface of the nutrient solution (Czopek 1959b) were considered to be fully germinated.

A thermostat with controlled light intensity (Fig. 1) was constructed for investigations on the germination in chromatic light. Four bulbs, 150 W each, and appropriate filters formed the source of blue and green light and four bulbs (60 W each) the source of red and far red light. These bulbs were cooled by means of a nearby placed ventilator. Two, 40 W, oblong bulbs placed in a closed chamber under the thermostat provided the heating system. The temperature was controlled by means of a contact thermometer and a mercury relay. Air circulation was assured by an small ventilator placed inside the thermostat chamber.

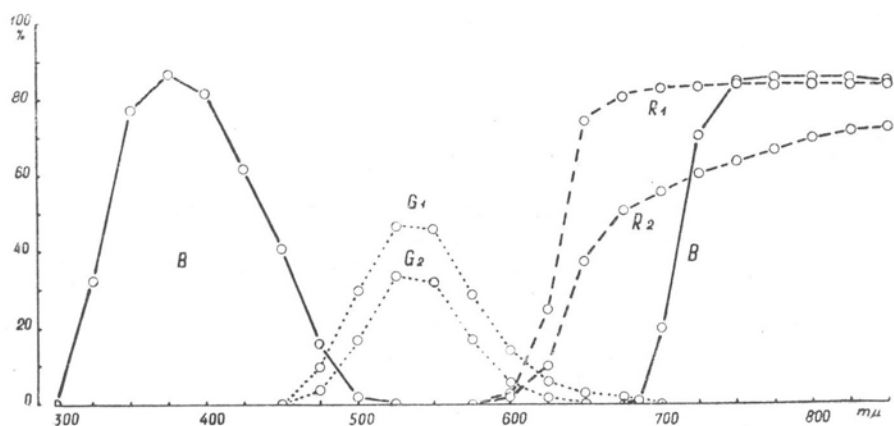


Fig. 2. Transmission spectra of the glass filters. Abscissae — wave-length in  $m\mu$ , ordinates — percentage of transmitted light. B — blue filter,  $G_1G_2$  — green filters,  $R_1R_2$  — red filters

Before reaching the chamber, the light passed through two kinds of filters: first through a liquid filter and then through glass filters. A specially adapted water container — 10 cm high — filled with a solution of an anorganic salt constituted the liquid filter. The upper surface of this solution was in contact with a glass plate. The coloured

glass filters characterized by strictly determined light transmissions were placed under the water container. A board equipped with switches enabled to set in motion the different parts of the apparatus. The spectral regions given by this arrangement (Fig. 1) varied in dependence on the filters. Following filters were used: one blue filter, 2 green ones, 2 red ones and 1 blue and 1 red filter for far red. The light transmission curves of the glass filters were determined by means of a spectrophotometer of Uvispek-Hilger type and are shown in Fig. 2.

The transmission range of the glass filters was narrowed by suitable liquid filters obtained by dissolving a mineral salt in 1 liter of a solution sulphuric acid (Fig. 3). The following salts and concentrations

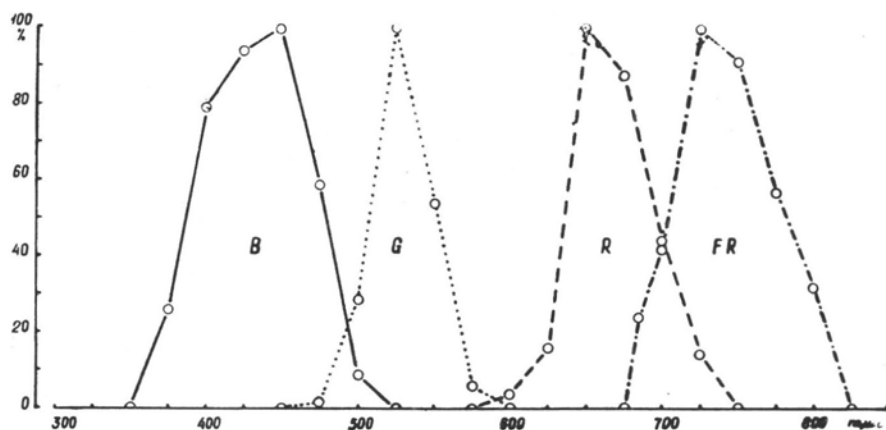


Fig. 3. Relative transmission data of the filter sets for different spectral regions. The transmission of the set is the result of the transmission of a) the glass filters, b) the liquid filter and c) of the spectral composition of the light source (Kliesznin 1954). Abscissae — wave-length in  $m\mu$ , ordinates — percent relative transmission. Spectral regions: B — blue; G — green; R — red; FR — far red

were used: a) for blue and green light: 50 g of copper sulphate in 0.5% sulphuric acid; b) for the red light: 300 g of ferrous ammonium sulphate in 2% sulphuric acid; c) for the far red light: 150 g of ferrous ammonium sulphate in 2% sulphuric acid (Withrow and Price 1953, Withrow and Elstad 1953). In all cases the liquid filters were 10 cm thick.

The intensity of the transmitted light corresponding to the different spectral regions was measured in absolute units ( $\text{ergs/cm}^2/\text{sec.}$ ) by means of a Kipp and Zonen thermopile and a galvanometer. An attempt was made to obtain nearly equal light intensities in all the spectral regions applied in our research.

Counts of germinated turions were performed in green light. The circumstance that the dark adapted eye is very sensitive to green light of about 510 m $\mu$  permitted to count the germinated turions in flasks exposed to green light of very low intensity. It is well known that green light in the region 500—550 m $\mu$  exerts a minimal action on photo-periodism, synthesis of chlorophyll and photomorphogenesis (Withrow and Price 1957). The fact however that green light promotes the germination of turions (as it is shown in further experiments) induced us to examine how far the light applied during the counting operation could modify the results of turion germination in darkness. It was observed that green light of low intensity applied 10—15 sec. daily caused a small rise of the germination rate (about 2%). This rise, however, was so small that it need not be taken into consideration.

Table 1  
Intensities and spectral regions of the applied chromatic light

Light	Intensity in ergs/cm <sup>2</sup> /sec.	Spectral regions m $\mu$		
		from	maximum	to
Blue	224,3	350	450	525
Green	217,2	450	525	600
Red	221,5	575	650	750
Far red	225,6	675	725	825

The experimental results are expressed in percent of germinated turions. In the final stage the differences between the replications do not exceed  $\pm 3\%$ .

## RESULTS

### 1. The germination of turions in dependence on the length of the illumination period

As it results from the former experiments on the germination of turions (Czopek 1959b) light plays the part of an important oligodynamic factor. In order to examine more detailfully the influence of small doses of light, turions were allowed to germinate in darkness and were then exposed to white light (intensity 10 660 ergs/cm<sup>2</sup>/sec.) every day for a short time. Fig. 4 shows the results of germination of turions illuminated 2, 5, 10 and 30 minutes daily and of turions exposed to continuous light or kept in darkness. A 2 minutes long exposition to

light already promotes the germination rate and the percent of germinated turions illuminated for 10 minutes daily is the same as in continuous light (Fig. 4).

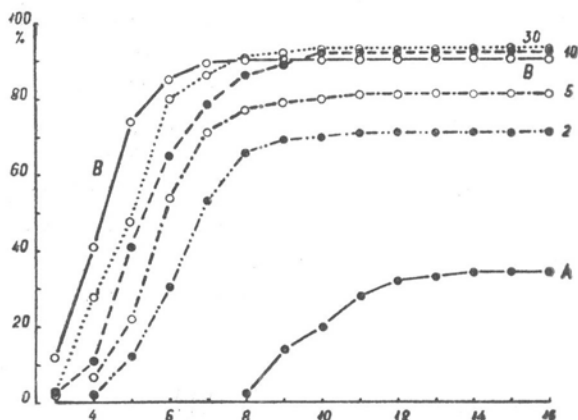


Fig. 4. Germination of turions in dependence on the length of illumination with white light. Abscissae — days, ordinates — germination percentage. A — controls (darkness); B — controls (continuous light), 2, 5, 10 and 30 daily times of illumination in minutes

## 2. Germination of turions maintained in darkness and subsequently exposed to light

In darkness the percentage of germinated turions is relatively low. The maximal value attained is about 35% and it did not undergo any further change. In order to ascertain that the ungerminated turions possess still the ability to germinate they were exposed to continuous

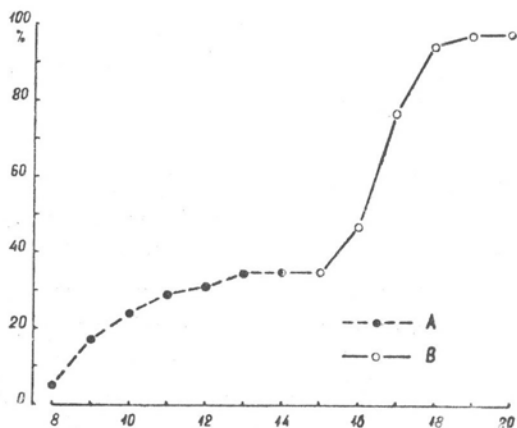


Fig. 5. Germination of turions in the dark period (A) and after transferring into light (B). Abscissae — days, ordinates — germination percentage

fluorescent white light of 10 660 ergs/cm<sup>2</sup>/sec. intensity. Turions were transferred to a light thermostat after 14 days of germination in darkness, when the maximal germination percentage was already attained. Fig. 5 shows that turions after being exposed to light, began to germinate very intensively and their germination percent attained 98. Evidently light is a factor stimulating the germination process, though, for a part of the turions it is not indispensable.

### 3. Germination of turions and light dosage

All the turions received the same quantity of light. In some series, however the whole quantity was supplied at one time, in others in daily doses of the same intensity but of proportionally shorter time. Ten minutes long daily irradiations applied for 6 days proved to be more advantageous than one 60 minutes long application of light on the 3-rd day of germination. When a daily 10 minutes long illumination was applied the turions germinated almost at the same rate as in continuous light, whereas a single but greater dose of light was followed by a lower percentage of germination (Fig. 6). Turions which received daily 5 two-minutes long doses of light (separated by 2 hours intervals) manifested only a little higher germination percentage (Fig. 6) than turions illuminated daily once for 10 minutes. Thus, the conclusion is that short daily doses are more advantageous for the germination process than a greater but single dose of light. The way light is applied during the day (one dose or several small doses) is of no importance for germination.

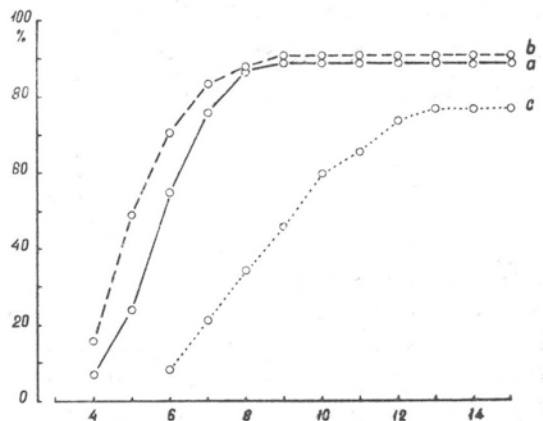


Fig. 6. Germination of turions in dependence on the dose of light. Abscissae — days, ordinates — germination percentage. *a* — 10 minutes daily; *b* — 10 minutes daily, 2 minutes long doses every 2 hours; *c* — single dose of light for 60 minutes on the third day

#### 4. Light doses applied in various phases of germination

If light is applied only once for 1 hour on the second day the germination rate and percentage are low. Illumination on the 4-th day stimulates germination and induces a higher germination percentage. A still stronger effect is obtained by applying light on the 8-th day, i.e.

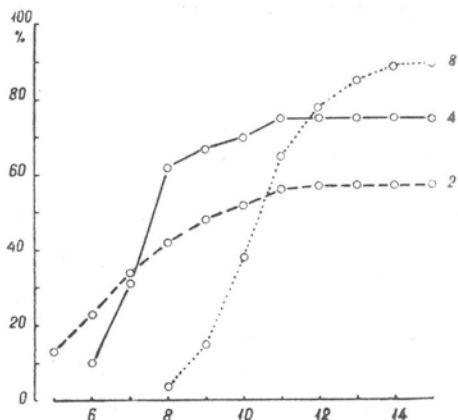


Fig. 7. Germination of turions exposed for 1 hour to a single dose of light. Abscissae — days, ordinates — germination percentage. 2, 4, 8 light doses on the second, 4<sup>th</sup> and 8<sup>th</sup> day respectively

the moment the germination in darkness has already started. Thus, the influence of a single dose of light is most effective if it is applied at the time the germination process has already started (Fig. 7).

Turions illuminated for 10 minutes on the first, second and third day i.e. till the beginning of germination, continue well to germinate in darkness, even if no light is applied further on. The germination

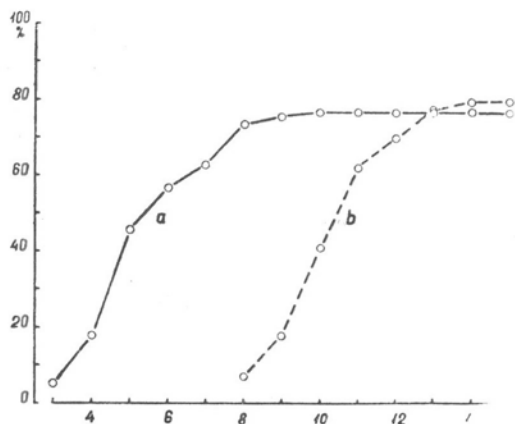


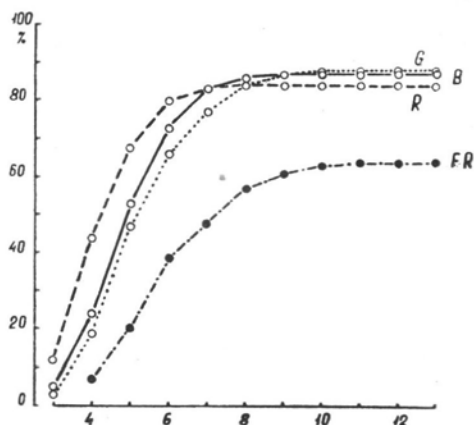
Fig. 8. Germination of turions illuminated 10 minutes daily. Abscissae — days, ordinates — germination percentage, a — daily illuminations till the 3<sup>rd</sup> day; b — from the 8<sup>th</sup> day

rate of turions exposed to light from the 8-th day on, for 10 minutes every day was almost equal to rate of turions which received one 60 minutes long dose of light on that day (Fig. 8). It may be supposed therefore that turions are at that time specially photosensitive and a single light dose is sufficient to assure light saturation.

## 5. Germination of turions in continuous light of various spectral regions

The turions were allowed to germinate at 28°C in a thermostat in continuous chromatic light. The spectral regions of the blue, green, red and far red light were specified in the methodical part. The intensity of chromatic light, about 223 ergs/cm<sup>2</sup>/sec., was relatively low in comparison with the white light (about 10 660 ergs/cm<sup>2</sup>/sec.) used in previous experiments. In all spectral regions light of almost equal intensity (Table 1) was applied. The results of these experiments show that turions are relatively sensitive for blue, green and red light. In no case, however, the germination percentage exceeded 90% (Fig. 9). Tu-

Fig. 9. Germination of turions in continuous chromatic light. Abscissae — days, ordinates — germination percentage. B — blue, G — green, R — red, FR — far red light



rions irradiated with far red begin also to germinate but somewhat later, viz. on the 4-th day. The percent of germinated turions does not exceed, however, 65%. Thus, a considerably lower germination percentage, in comparison to other spectral regions, was observed in this case, although the far red contained a certain amount of red rays (Fig. 3). The curves representing the germination percentages in blue, green and red light are similar in shape. For this reason it is impossible to determine more precisely the spectral sensitivity of turions.

## 6. Action of short irradiations with chromatic light on the germination of turions

In these experiments the germination of turions in darkness was interrupted by 10 minutes long irradiations with chromatic light applied always at the same time during several days. In red light turions began to germinate on the 5-th day, in blue and green on the 6-th day (Fig. 10); the percentage, however of germinated turions did not exceed 70%. In far red light germination started on the 7-th day and the final

percentage of germination was considerably higher than that of the controls kept in darkness. This is most probably connected with the presence of a small quantity of red rays in the spectral region transmitted by the far red filter. An analysis of the graphs suggests the conclusion that the turions are very photosensitive; they respond even to light delivered in small doses of low intensity. If germination pro-

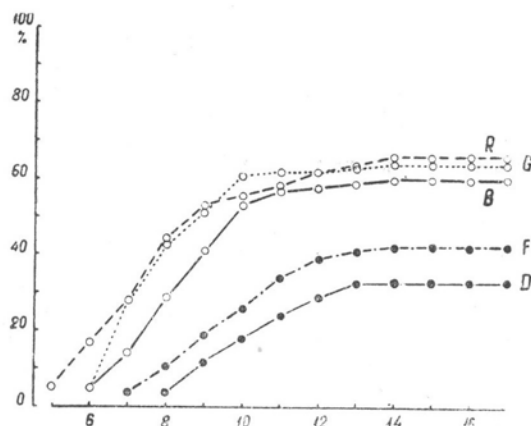


Fig. 10. Germination of turions exposed for 10 minutes daily to chromatic light. Abscissae — days, ordinates — germination percentage. B — blue; G — green; R — red; FR — far red light; D — dark control

ceeds in darkness and is interrupted every day for 10 minutes by blue, green or red light, its rate is much enhanced and a much higher percentage is attained in comparison with germination in continuous darkness. Far red light stimulates germination as well, but its action is much weaker.

#### 7. Photoreversibility red — far red

The reversibility of the action of red and far red light on the germination of turions made the subject of a special study. The turions were first allowed to germinate in darkness and then were illuminated every day 10 minutes with red and 10 minutes with far red light. The far red light was applied either immediately after the action of red light or after a delay of 3 hours. Moreover, the illumination effects of the sequences red — far red — red and red — far red — red — far red were also examined.

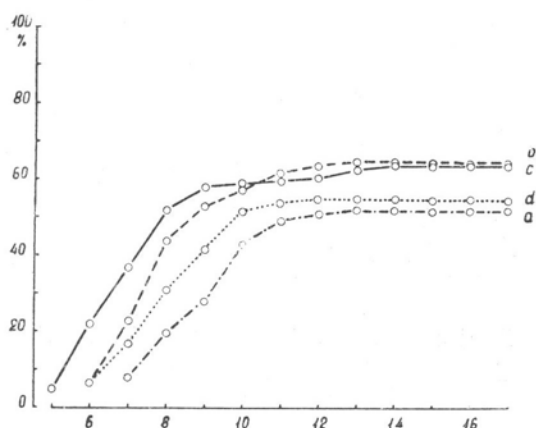
The stimulating action of red light on the germination was partly reversed by an immediate treatment with far red light. The percentage of germination is far lower than for red light but higher than for far red light only (Fig. 11).

If, however a delay of 3 hours is introduced between the times of application of red and far red radiations the final effect will be the same as for red light.

If in the sequence of lights: red — far red — red, one kind of radiation is applied immediately after the other, the final effect is the same as if the turions were exposed to red light only.

On the contrary an immediate succession of red — far red — red — far red radiations leads to a somewhat higher germination percentage than for far red alone, but considerably lower than for red light (Table 2).

Fig. 11. Germination of turions illuminated with red and far red light, each for 10 minutes every day. Abscissae — days, ordinates — germination percentage. Light sequences: a) red — far red (immediately); b) red — far red after 3 hours; c) red — far red — red (immediately); d) red — far red — red — far red (immediately)



These results suggest that the final effect of light on the germination process is determined by the kind of light applied in the last instance, provided that the different kinds of light are applied in immediate succession. Far red light applied 3 hours after red light does not inhibit the germination process which proceeds like after red.

Table 2

Germination of turions in dependence on the illumination with red and far red light

Light doses — 10 minutes daily	Germination percentage
Darkness (control)	33
Red	66
Far red	42
Red — far red	52
Red — far red after 3 hours	66
Red — far red — red	64
Red — far red — red — far red	55
Far red — red	70
Far red — red — far red	55

If however, the irradiation with far red light is followed immediately by red light the germination will proceed like for red. On the contrary immediate illumination with far red — red — far red lights gives

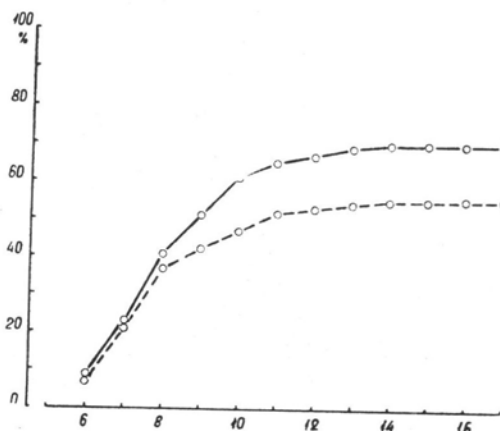


Fig. 12. Germination of turions illuminated with far red and red light, each for 10 minutes every day. Abscissae — days, ordinates — germination percentage. Light sequences: a) far red — red (immediately); b) far red — red — far red (immediately)

a final effect like for germination in far red. Thus, the inhibitory effect of far red light is reversed by irradiation with red light (Fig. 12).

#### 8. Photoreversibility of the light sequences: blue — far red and green — far red

In order to examine the photoreversibility of the light sequences: blue — far red and green — far red similar experiments as with far red light were carried out with blue, green and far red radiations. It results from former experiments that blue light stimulates the germination of turions. An immediate exposure to far red light after blue light leads to a decrease of the germination percentage, whereas, the

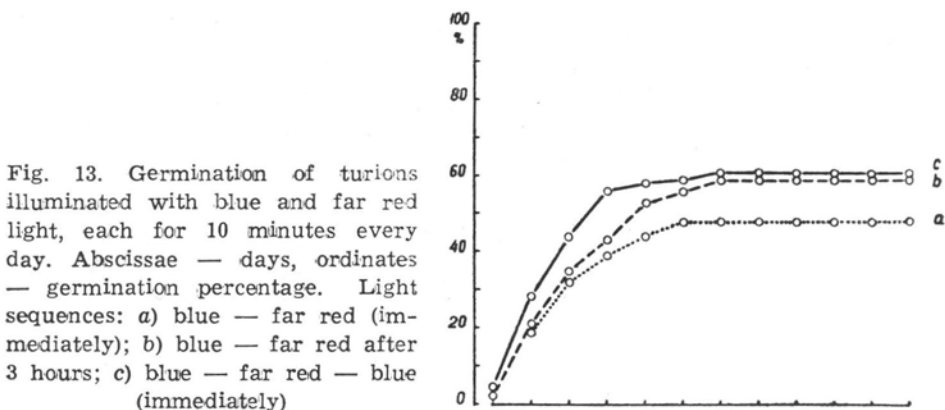


Fig. 13. Germination of turions illuminated with blue and far red light, each for 10 minutes every day. Abscissae — days, ordinates — germination percentage. Light sequences: a) blue — far red (immediately); b) blue — far red after 3 hours; c) blue — far red — blue (immediately)

treatment with far red after a delay of 3 hours is without action on germination. An immediate succession of blue — far red — blue radiations gives a similar effect to that obtained after illumination with blue light only (Fig. 13).

Table 3

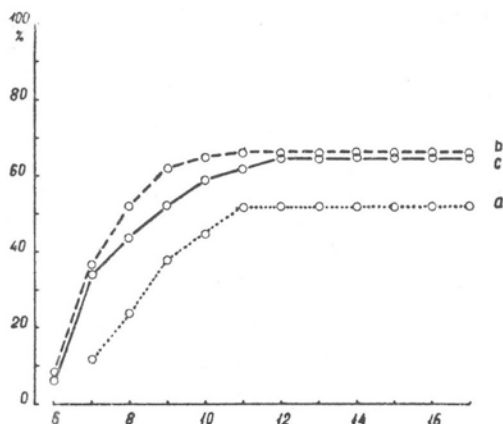
Germination of turions in dependence on the illumination with blue — far red and green — far red light

Light doses — 10 minutes daily	Germination percentage
Blue	60
Blue — far red	48
Blue — far red after 3 hours	59
Blue — far red — blue	61
Green	64
Green — far red	52
Green — far red after 3 hours	66
Green — far red — green	65

Germination is also stimulated by green light. Its action can be reversed by an immediate application of far red light. A 3 hours long delay in the application of far red radiation is without notable effect on the germination percent. The following sequence of lights: green — far red — green exerts a final effect similar to that obtained with green light only (Fig. 14).

The results of these experiments suggest the conclusion that the final effect of the action of chromatic light on the germination of turions is determined by the kind of light used in the last instance, on the condition that one kind of light is applied immediately after the preceding

Fig. 14. Germination of turions illuminated with green and far red light, each for 10 minutes every day. Abscissae — days, ordinates — germination percentage. Light sequences: a) green — far red (immediately); b) green — far red after 3 hours; c) — green — far red — green (immediately)



antagonistic kind of radiation. A delay of 3 hours in the application of far red light is sufficient to deprive this kind of light of its inhibitory action on the germination of turions.

#### DISCUSSION

The results of our experiments indicate that turions of *Spirodela polyrrhiza* are characterized by a high degree of photosensitivity. They are able to germinate in darkness, but the percent of germinated turions is low; it becomes notably increased even by a small dose of light. Thus light is an important oligodynamic (non trophic) factor. A similar oligodynamic action on germination was also observed in many photosensitive seeds. In this respect seeds of various species behave very differently. Seeds of some *Nicotiana* species belong to the most sensitive ones, since maximal germination is attained already after one minute long irradiation with 1000 lux (Isikawa and Shimogawara 1954, cit. acc. Borthwick and Hendricks 1961). In spite of considerable physiological and anatomical differences shown by seeds and turions (the way of their formation and their water content 61,8% fresh weight, Czopek 1959b) the latter have many properties in common with seeds and spores.

Light may exert a stimulating or inhibiting action on the germination of many seeds, although it is almost without any influence on the majority of seeds or spores. Mohr (1956) found that the spores of *Dryopteris filix-mas* germinate in light only. The germination in darkness of turions of *Hydrocharis morsus ranae* is stimulated only by the hormone 2,4 D (Wiśniewski 1958). In their experiments on germination of the seeds of *Nemophila insignis*, Black and Wareing (1957, 1960) established that continuous light depresses the germination rate, which in darkness attains 90%. Sircar and Biswas (1960) point out that white light stimulates the germination of rice seeds only at a very low percentage as they are also able to germinate in darkness perfectly well. Light influences the germination of turions of *Spirodela polyrrhiza* favourably. It is interesting to notice that an every day treatment with even small doses of light is much more effective than a single but more intensive dose. A single dose of light proves to be most effective when it is applied at the very beginning of germination in darkness. It seems that in turions the light absorption system is not able to utilize the total amount of a more intensive but single dose of light. It has been found, during the study on the action of various spectral regions on turion germination, that in continuous blue, green and red light similar results are obtained as in white light. Thus

it is impossible to establish with precision the spectral sensibility of germinating turions. Far red radiations also promote their germination, but the observed stimulation is much weaker in comparison with other kinds of chromatic light of the same energy. An inhibiting influence of blue light on germination was observed with fern spores (Mohr 1956), exposed to low light intensities ( $200 \text{ ergs/cm}^2/\text{sec.}$ ). Of all spectral regions, however, the far red region  $730\text{--}750 \text{ m}\mu$ , exerted the most inhibiting action, whereas red rays ( $650\text{--}670 \text{ m}\mu$ ) gave the best germination results (Mohr 1956). Jones and Bailey (1956) illuminated the seeds of *Lamium amplexicaule* with blue, green and red light for 6 minutes and observed only a slight decrease of the germination percentage. Black and Wareing (1957, 1960) found that the germination of *Nemophila* seeds was considerably inhibited by blue ( $452 \text{ m}\mu$ ) and far red ( $710 \text{ m}\mu$ ) and less by blue-green light ( $483\text{--}496 \text{ m}\mu$ ). In green ( $542 \text{ m}\mu$ ) and red light ( $651 \text{ m}\mu$ ) the inhibition was very low. Sircar and Biswas (1960) point out that the germination rate of rice seeds is depressed in comparison with darkness by red light only at a small degree, whereas, far red light inhibits it stronger. Evenari *et al.* (1957) noticed that germination of lettuce seeds is checked by blue light of maximal energy at  $430 \text{ m}\mu$  after a 5 minutes long irradiation, provided that it is applied within the first 6 hours following the beginning of imbibition. The same kind of light applied after 10 hours exerts a stimulating influence. Kadman-Zahavi (1960) established that blue light has no ability to initiate the germination of *Amaranthus retroflexus* seeds. It can be seen from the above mentioned facts that in most cases blue light does not stimulate the germination of seeds in contrast to turions which are particularly sensitive to short wave radiations. With the exception of turions no stimulating action on germination was observed with green light.

It was shown that the action of red and far red radiations on the germination of turions are antagonistic and reversible processes. The stimulating effect of red light on the germination was reversed to a large extent by an immediate treatment with far red light.

The presence of a photoreversible absorption system red—far red was discovered in many germinating photosensitive seeds. Flint and McAlister (1935, cit. acc. Wassink and Stolwijk 1956) described the stimulating influence of red light on the germination of lettuce seeds. Germination could be inhibited by the action of far red of about  $730 \text{ m}\mu$  maximal activity. The antagonistic influence of red and far red light on the germination process of lettuce seeds made the object of many studies (Borthwick *et al.* 1952, Liverman and Bonner 1953, Miller 1956, Toole *et al.* 1956, Wareing 1956,

Elliot and French 1959, Haber 1959, Skinner and Shive 1959, Kahn 1960a—b, Ikuma and Thimann 1960, 1961, Surrey 1961). The reversible red — far red reaction during the germination process was examined also in seeds of *Amaranthus retroflexus* (Kadman-Zahavi 1960), *Elsholtzia* (Isikawa and Ishikawa 1960), *Lamium amplexicaule* (Jones and Bailey 1956), *Lepidium densiflorum* and *Lepidium virginicum* (Toole et al. 1955, 1956), *Nemophila insignis* (Black and Wareing 1957, 1960), *Pinus virginiana* (Toole et al. 1961), *Oryza sativa* (Sircar and Biswas 1960).

If far red light is applied not immediately after the red light but after a delay of 3 hours, the final effect on the germination of turions is the same as for red light. Borthwick et al. (1954, cit. acc. Ikuma and Thimann 1960) established that far red light does not inhibit the germination of lettuce seeds if it is applied after a long delay following the treatment with red light. Ikuma and Thimann (1960) found that the inhibition effect of far red light decreases gradually with the increase of the time elapsing between the moments of irradiation with red and far red light. Far red radiation does not inhibit the germination of lettuce seeds when applied 8—9 hours after red light. Kadman-Zahavi (1960) noticed that the inhibition of germination of *Amaranthus retroflexus* seeds was most marked when far red light was applied after 2—16 minutes and not immediately after the former irradiation.

The sequence of lights: red — far red — red gives a final effect similar to that obtained for red light. Thus, the conclusion may be that the final effect on turions germination depends on the kind of light applied at the end, provided that the different kinds of light are administered immediately one after the other. Similar results were obtained for fern spores (Mohr 1956), lettuce seeds (Borthwick et al. 1952, Borthwick et al. 1954, cit. acc. Borthwick and Hendricks 1961, Haber 1959), *Lamium amplexicaule* (Jones and Bailey 1956) and *Pinus virginiana* seeds (Toole et al. 1961).

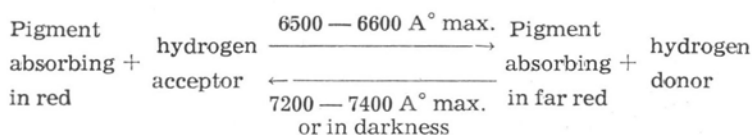
The inhibitory influence of the far red light on turion germination could be reversed by an immediate treatment with red light. This reversibility was not observed with seeds of *Elsholtzia* (Isikawa and Ishikawa 1960).

The promoting effect of blue and green light on the germination of turions could be reversed by an immediate application of far red light. It is supposed nowadays that the antagonistic effect of red and far red radiation is connected with the presence in the seeds of a specific pigment — phytochrome (Butler et al. 1959, Borthwick and Hendricks 1961). The results of our study permit to suppose that

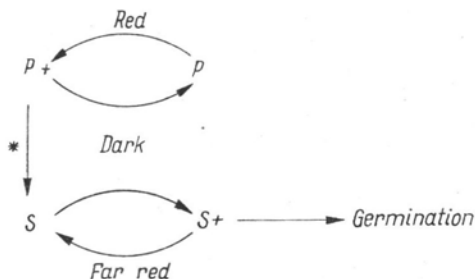
an analogous absorption system participates also in the germination process of turions. The phytochrome, however, present in turions has the ability to absorb blue and green radiations to the same extent as red light. It will be, however, a subject of further investigations.

Several hypotheses have been advanced to elucidate the antagonistic action shown by red and far red radiations on plants. Borthwick *et al.* (1952) suppose that the photochemical influence of light on seed germination is dependent on the presence of two pigments: the first absorbs red, the second far red light (absorption maxima about 650 m $\mu$  and 730 m $\mu$  respectively). Further investigations of these authors, however, have shown that these pigments are only two forms of the same compound. Butler *et al.* (1959), Borthwick and Hendricks (1961) obtained phytochrome *in vitro* from etiolated seedlings of *Zea mays*. The absorption curve of this pigment in the long wave spectral region is similar to the curve corresponding to the action spectra of photoperiodic flowering responses, germination of seeds, internode elongation *etc.*

It is assumed that a reversible photoreaction red — far red is controlling the cellular metabolism and involves the interconversion of two photoreceptors (Borthwick and Hendricks 1961). According to Borthwick and Hendricks (1961) the photoreaction can be written:



On the contrary, the action of red and far red light on the seed germination is explained by Kadman-Zahavi (1960) by means of the following scheme:



By a short illumination with red light the pigment  $P$  is converted into an activated form  $P+$  which in a dark reaction acts on a com-

\* the arrow in the scheme was added by the present author.

pound  $S$ . In turn  $S$  is converted into an activated form  $S^+$ ; at the same time  $P^+$  returns to its original state  $P$ . The substance  $S^+$  in the activated state stimulates germination. If however  $S^+$  is illuminated by far red light, it is converted back into  $S$ , and becomes incapable of promoting germination. The reversibility of the reaction results from the fact that the substances involved in the reaction are at the end in their original states  $P$  and  $S$  (Kadman-Zahavi 1960).

As it has already been established far red light does not inhibit the germination of turions if the time elapsing between treatments with red and far red light exceeds 3 hours. By combining the above mentioned results with the hypothesis advanced by Kadman-Zahavi (1960) it could be supposed that during this time (3 hours) a sufficient amount of substance  $S^+$  necessary to stimulate germination was produced. The substance  $S^+$  in the excited form activates the metabolic enzymes and then reverts to its original state. In these conditions far red light is without any influence because there is no substance — in an activated form  $S^+$  — which would be sensitive to far red radiation. Further investigations will show how far this supposition is experimentally substantiated.

#### SUMMARY

This paper presents the principal results of a study on the action of different spectral regions (blue, green, red and far red) on the germination of turions of *Spirodela polyrrhiza*.

1. The germination of turions exposed to continuous white light attains 100%. In darkness the percent of germinated turions is much lower (about 35%), but a daily several minutes long dose of light is sufficient to induce 100% germination.

2. Turions exposed to continuous white light or kept in darkness begin to germinate on the 3<sup>rd</sup> or 8<sup>th</sup> day respectively.

3. If the turions are allowed to germinate in darkness, their sensibility for light increases and attains a peak value the moment they begin to germinate (after 8 days).

4. The percentage and rate of germination of turions exposed to continuous blue, green and red light are the same as of turions germinating in white light. On the contrary, far red light exerts an inhibitory action on the germination process.

5. The stimulating effect of red light could be reversed by treatment with far red radiation, provided that it was applied immediately after red light.

6. If the turions are exposed in succession to different chromatic light (for inst. red — far red — red light), the final effect on the germination is determined by the kind of light administered in the last instance, provided that this light is supplied immediately after the precedent light.

7. If a delay of about 3 hours is included between the moments of irradiation with red and far red light, the far red light is without inhibitory action on the germination of turions.

8. The inhibitory effect of far red light could be reversed by an immediate irradiation with red light.

9. A short irradiation of turions with blue or green light promotes their germination. This effect can be reversed by a subsequent immediate illumination with far red light.

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