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Photochemical activity of chloroplasts of isogenic lines (*E* genes) of soybean (*Glycine max* (L.) Merr.) under different periods of red-light irradiation

V.F. Timoshenko, V.V. Zhmurko

The effect of different periods of red-light (RL, 660 nm) irradiation of plants on the biomass, leaf area, chlorophyll content, level of the Hill reaction and photophosphorylation in short-day (SD) and photoperiodical neutral (PhPN) lines of soybean have been studied in vegetation experiments. The objects of study were the isogenic lines (*E* genes) of soybean (*Glycine max* (L.) Merr.), Clark variety. The SD (*E1E2E3*) and PhPN lines (*e1e2e3*) were used. The plants were grown in a vegetation chamber in the soil culture (black soil). Plants of SD and PhPN lines were grown in 9 vessels of three liters volume. 10–12 plants were grown in each vessel. The constant growing conditions were provided during the experiment: temperature – 20–24/17–20°C (day/night), soil moisture – 60–70 % of the total soil moisture content, intensity of illumination – 20 klx, photoperiod duration – 10 hours. After 4–5 weeks of vegetation (after the second true leaf formation), plants of each line in three vessels were irradiated for 30 minutes with low intensity red light at the beginning (experiment 1) or in the middle of the dark period (experiment 2). The light diodes emitting in the region of 630±10 nm were used for plant irradiation. Other plants in three vessels of each line, which were not illuminated by the red light, were used as a control group. It was shown that under the short day the SD line in the control group passed to flowering 43±1.8 days after germination and PhPN line – 44±2.2 days after germination. The red-light irradiation, both before the beginning and in the middle of night, caused a delay of the transition to flowering in the SD line by 5±1 and 7±2.2 days, respectively. In the PhPN line, changing flowering period due to RL was not established. In the SD line, activation of the phytochromes by RL before the dark period caused an increase of the biomass, leaf area, total chlorophyll content, reduction of potassium ferrocyanide and photophosphorylation by isolated chloroplasts per chlorophyll of one leaf. While interruption of the night by RL caused decrease of these parameters and Hill reaction intensity per 1 mg of chlorophyll. The effect of RL on the studied parameters in the PhPN soybean line has not been detected.

Key words: soybean *Glycine max* (L.) Merr., isogenic lines, phytochrome, photochemical activity of chloroplasts, morphogenesis.

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Introduction

In the process of plant ontogeny, the changes occur in the formation of their attracting organs. Plant growth, development and photosynthesis are changed by environmental factors, of which light is very important (Voskresenskaya, 1987). The main regulator of morphogenesis, which affects the vast majority of metabolic processes, is the phytochrome system. Phytochromes are activated by red light (RL) with a wavelength of 660 nm, while light of 730 nm translates the phytochrome into an inactive form (Qvail, 2010). The literature provides data of many physiological effects of RL, one of which is related to the carbohydrate metabolism of plants. It was shown that interruption of the dark period by RL in the short-day photoperiodic cycle caused a significant decrease of the outflow of carbohydrates from the leaves (Tsybulko, 1998). According to the author, this is one of the reasons of a delay of the transition to flowering of short-day plants under short photoperiod. It was also shown that in the afternoon, RL irradiation caused a change in the dynamics of carbohydrate content (Timoshenko, Zhmurko, 2013) and the activity of sucrose phosphate synthase, which is one of the key enzymes of sucrose synthesis (Shchegolev, Zhmurko, 2013).

In experiments with winter wheat varieties manifested long-day (LD), short-day (SD) and photoperiodical neutral (PhPN) reactions to the photoperiod duration, it was shown that during the interruption of the long dark period in the SD photoperiodic cycle, the LD varieties accelerated the transition to spinning, SD delayed it and PhPN did not respond to such influence (Zhmurko et al., 1997). At the same time, LD significantly increased the outflow of carbohydrates from the leaf, and SD significantly decreased (Zhmurko, 2009).

Under conditions of an interruption of the dark period, the activity of oxidoreductases and ATPase increased in the SD variety, but in the LD variety it decreased (Zhmurko, 2009).

Therefore, in plants of winter type of development, effects are manifested from the activation of phytochromes. The effects are similar to those of plants of the spring type of development of different photoperiodic groups.

The above also indicates that an activation of phytochromes affects the metabolism of carbohydrates of plants of different photoperiodic groups.

To determine the role of the phytochrome system in regulating the process of plant transition to flowering and in the photoperiodic reaction, it is important to study the effect of phytochromes on the photosynthetic characteristics. According to Tsybulko, the role of photosynthesis in the photoperiodic response of plants is important, but there is no consensus about the presence and nature of such a link among scientists (Tsybulko, 1998). In addition, photosynthesis was generally considered as the process of formation of organic compounds. However, primary photosynthesis reactions provide energy, along with secondary photosynthesis reactions, other physiological and biochemical processes. In particular, photosynthesis supplies energy and plastic substances for forming processes in apices of shoots during the photoperiodic reaction of plants.

The question of the possible involvement of photoperiodic sensitivity genes in the photosynthesis regulation under the activation of phytochromes remains unknown, although these genes play an important role in the initiation of flowering (Chincinska et al., 2008; Matsoukas et al., 2012).

From this point of view, in order to study this issue it is advisable to use the near isogenic lines of plants, carrying genes of photoperiodic sensitivity control, as a plant material. In particular, these may be soybean plants, in which the genes that control the time from germination to flowering (*E* genes) have been identified. Today, in soybean 8 major early maturity *E*-series genes have been identified that control time to flowering and ripening: *E1*, *E2*, *E3* (Buzzell, 1971), *E4* (Buzzell, Voldeng, 1980), *E5* (McBlain, Bernard, 1987), *E6* (Bonato, Velio, 1999), *E7* (Cober, Voldeng, 2001) and *E8* (Cober et al., 2010), as well as the locus *J* (long juvenile). In the presence of the last locus, soybean plants bloom late even under the short day (Ray et al., 1995).

According to the above, the purpose of our research was to study the effect of different activation periods of phytochromes by RL (660 nm) on the biomass, leaf area, chlorophyll content, Hill reaction and photophosphorylation in the SD and PhPN isogenic lines of soybean (*Glycine max* (L.) Merr.).

Materials and methods

The objects of study were isogenic lines (*E* genes) of soybean (*Glycine max* (L.) Merr.) Clark. We used the SD line (*E1E2E3*) and PhPN line (*e1e2e3*). The seeds of the lines were obtained at the National Center for Plant Genetic Resources of Ukraine and were reproduced at the experimental site of the Department of Plant and Microorganisms Physiology and Biochemistry of V.N.Karazin KhNU. Since soybeans are self-pollinated, the genetic homogeneity of the lines has been maintained by control to prevent mechanical mixing of the seeds of the different lines during harvesting.

The plants were grown in the vegetation chamber of the Department of Plant and Microorganisms Physiology and Biochemistry of V.N.Karazin KhNU in the soil culture (black soil). The SD and PhPN plants were grown in 9 vessels of three liters volume. 10–12 plants were grown in each vessel. The constant growing conditions have been provided during the experiment: temperature – 20–24/17–20°C (day/night), soil moisture – 60–70 % of the total soil moisture content, intensity of illumination – 20 klx, photoperiod duration – 10 hours. After 4–5 weeks of vegetation (after the second true leaf formation), plants of each line in three vessels were irradiated for 30 minutes with low intensity of red light at the beginning (experiment 1) and in the middle of the dark period (experiment 2). The light diodes emitting in the region of 630±10 nm were used for plant irradiation. Other plants in three vessels of each line, which were not illuminated by the red light, were used as a control group.

A fully developed second leaf from the apex was taken into the analysis. Biomass and leaf area were evaluated after sevenfold irradiation. Sampling for the analyzes of the photochemical activity of isolated chloroplasts was performed at 9.00 and 13.00 after fourfold and sevenfold irradiation of plants.

The content of chlorophyll was determined by spectrophotometric method after extraction of chlorophyll with 80 % acetone at a wavelength of 663 and 645 nm. The photochemical activity of chloroplasts (Hill reaction) was determined by the rate of reduction of potassium ferricyanide, photophosphorylation – by reducing the content of inorganic phosphate in the reaction mixture of the Hill reaction (containing KH_2PO_4 and ADP) (Gavrilenko et al., 1975). PA (Pure for Analysis) reagents were used for the analyzes. The leaf area was determined using the program "Photo M".

The significance of differences was calculated using the Student's *t*-test (Dospekhov, 1972). The tables present average values of 6–8 definitions.

Results and discussion

Phenological observations made it possible to establish that plants of the control group of two soybean lines bloomed almost simultaneously. Under the short day, SD plants bloomed 43 ± 1.8 days after germination. PhPN plants bloomed 44 ± 2.2 days after germination. As for the effect of red light at the flowering period, in both cases, both irradiation before beginning of the dark period and interruption of the night by red light, caused a delay of the transition to flowering of SD plants. Under RL irradiation before beginning of the dark period, the delay was 5 ± 1.0 days and under irradiation in the middle of night, the SD line bloomed 7 ± 2.2 days later than the control plants. The effect of RL on the change of flowering period in the PhPN line was not detected.

Studying the effect of RL on photosynthetic processes, we began by determining the change of biomass after irradiation of plants. Fig. 1 shows the values of dry biomass of control plants and irradiated plants. In the SD line, the result of exposure to the red light depended on the period of plant irradiation. Irradiation before the start of dark period accelerated the biomass growth. After seven exposures it was greater than the control by 15%. Whereas, after interruption of the night by RL, it was found a decrease of the biomass of irradiated plants compared to the control (after seven illuminations – by 13%). We have not established a biomass change of the PhPN line under the RL influence. Shpilova and Shchegolev observed earlier the varietal differences of growth reaction of tomatoes under the RL influence. Thus, during week after the termination of the RL irradiation, the plant biomass of the early Kremenchuk variety, breeding by the Ukrainian Institute of Vegetables and Melon Growing, continued to grow relatively to control. The changes for the late Ace 55 vf variety (Asgrow company) were not observed (Shpilova, Shchegolev, 2008).

The decrease of biomass that we observed in the SD soybean plants after interruption of night by RL may be explained by the fact that a long dark period is required for the complete outflow from photosynthetic organs of assimilates. An interruption of the dark period can disrupt starch hydrolysis in chloroplasts at night and outflow of carbohydrates from leaves (Tsybulko, 1998). A disruption of outflow, in turn, can cause a deficit of support of plastics and energy equivalents required for growth processes.

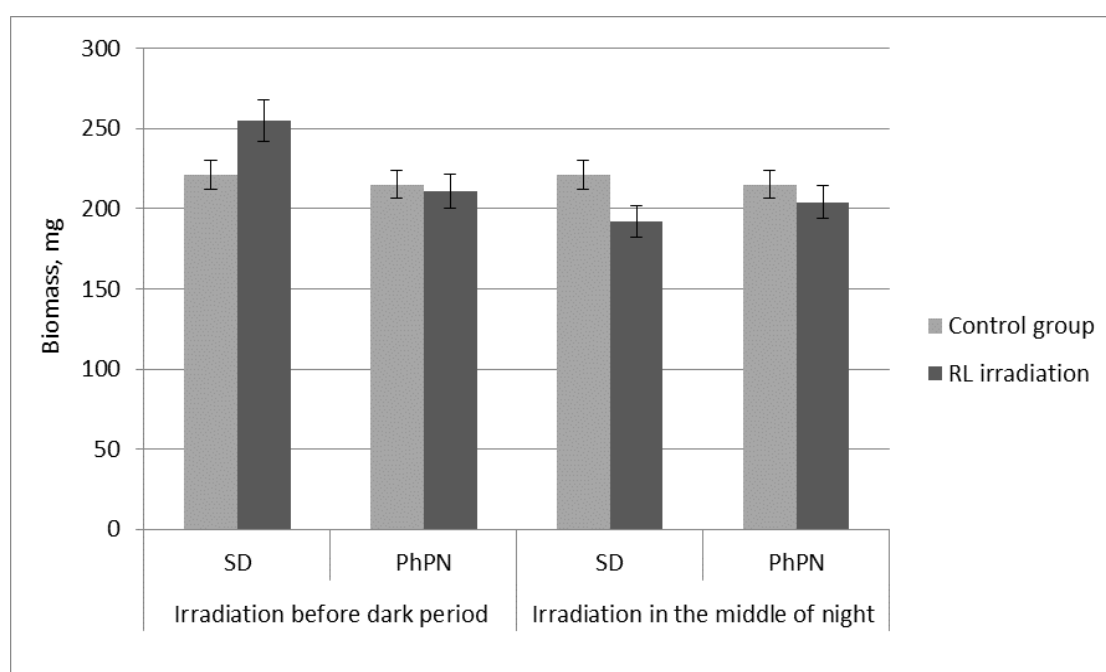


Fig. 1. Effect of RL on a biomass of the SD and PhPN soybean lines under different times of RL irradiation

It is known that a support of growth with assimilates determines the photosynthetic apparatus and processes of transport of assimilates to the attracting organs (Martirosyan et al., 2013; Tsybulko, 1998). For studying the productivity of photosynthesis, the indicators characterizing the size of the photosynthetic apparatus and the intensity of its functioning should be estimated. In response to RL, we consider extensive indicators including an area of the leaves as the main photosynthetic organs and content of chlorophyll in leaves; the indicators of the intensity of photosynthetic apparatus including the rate of photochemical reactions.

The study of the total area of leaves (table 1) showed that in the SD line the RL irradiation before the dark period caused the growth of photosynthetic surface. An interruption of night by the illumination of RL, in contrast, led to a decrease of the size of the total leaf area compared with control plants.

In addition, we analyzed effect of RL on an area of the second leaf from the apex. Because, as we know, the second leaf is fully formed and has the highest photosynthetic activity (Mokronosov, Gavrilenko, 1992). We used it for physiological and biochemical analyzes. The analysis of the second leaf area of the SD line showed the same directionality of changes under the RL influence. However, larger difference of its size between the control and irradiated plants was shown than of the total area of leaves. Thus, the second leaf area of SD plants irradiated with RL before night was 13 % higher than in the control, and this difference was 7 % for the total area. Under an interruption of night by RL, the reduction of the area of second leaves relatively to the control was 16 % and of the total area of leaves – 10 %. This pattern may be due to the fact that second leaves from the apex are formed when the RL irradiation has been carrying out.

As for different effects of the evening and night RL irradiation on growth processes, we believe that the stimulating effect of evening RL irradiation on the SD line can be considered as a reaction of short-day plants to increase the duration of photoperiod, which is manifested in the vegetative growth and delay of development. The decrease of leaf surface as a result of an interruption of night by RL can be a result of metabolic disorders (Tsybulko, 1998).

The effects of RL irradiation on the area of leaves of the PhPN line were not observed in our experiments.

Table 1.
Leaf surface area of the SD and PhPN isogenic soybean lines (cm²) under the RL irradiation

Variant	The total leaves area		The second leaf area	
	Exposure time			
	Before dark period	In the middle of night	Before dark period	In the middle of night
SD line, genotype $E_1E_2E_3$				
Control group	96.3 ± 4.8	96.3 ± 4.8	32.6 ± 1.6	32.6 ± 1.6
RL irradiation	102.8* ± 5.2	85.7* ± 5.0	35.9* ± 1.5	29.2* ± 1.4
PhPN line, genotype $e_1e_2e_3$				
Control group	95.4 ± 5.1	95.4 ± 5.1	29.4 ± 1.6	29.4 ± 1.6
RL irradiation	96.2 ± 14	96.1 ± 13	30.7 ± 1.7	28.5 ± 1.4

* differences with control are significant: $P \leq 0.05$.

One of the important criteria of evaluating the plant photosynthetic potential is the content of photosynthetic pigments. The determination of the content of main photosynthetic pigments chlorophylls showed (table 2) that in the SD plants the RL irradiation before the dark period led to an increase of the content of chlorophyll sum in leaves compared with the control plants irradiated four and seven times. An interruption of night by the RL irradiation in this line caused a decrease of the chlorophyll content. In PhPN plants effects of RL on the content of chlorophylls were not detected either in the case of evening irradiation or after night irradiation.

The stimulating effect of RL on the chlorophyll content was observed by other authors (Golovatskaya et al., 2012), when the RL irradiation had been conducting during the light period. RL decreased the lag-phase of the chlorophyll synthesis and increased its content. We obtained a similar result in the SD line under the RL irradiation at the end of the light period. But during the interruption of night by RL in the SD line there was a decrease of the chlorophyll content. This can be explained as

follows. An interruption of the night, according to Tsybulko, disrupts the hydrolysis of starch in chloroplasts (Tsybulko, 1998). Carbohydrates are also known to be a source of carbon skeleton for the synthesis of organic compounds including chlorophyll. It is likely that disruption of starch hydrolysis after the interruption of night by RL resulted in a decrease in the chlorophyll synthesis.

Table 2.
Influence of different RL irradiation terms (before dark period and at night) on the content of chlorophyll sum in leaves of the SD and PhPN isogenic lines

Variant	Number of exposures	Chlorophyll content, mg/g wet weight	
		Before dark period	In the middle of night
SD line, genotype $E_1E_2E_3$			
Control group	–	1.01 ± 0.05	1.01 ± 0.05
RL irradiation	4	1.12* ± 0.05	0.83* ± 0.06
Control group	–	1.03 ± 0.04	1.03 ± 0.04
RL irradiation	7	1.21* ± 0.06	0.76* ± 0.06
PhPN line, genotype $e_1e_2e_3$			
Control group	-	0.98 ± 0.04	0.98 ± 0.04
RL irradiation	4	1.02 ± 0.06	0.96 ± 0.05
Control group	-	1.02 ± 0.05	1.02 ± 0.05
RL irradiation	7	1.06 ± 0.06	0.94 ± 0.06

* differences with control are significant: $P \leq 0.05$.

The central process of primary photosynthesis reactions is a transport of electrons in the electron transport chain (ETC) of chloroplasts. The Hill reaction is the model reaction that characterizes the electron transfer intensity through ETC. This reaction has similar dynamics of changes under the influence of external factors with photosynthesis as a whole (Mokronosov, Gavrilenko, 1992). The results of the analysis of the RL influence on the Hill reaction are presented in table 3. Since the photochemical activity of chloroplasts is a dynamic indicator, we have evaluated the reaction twice a day: in the morning – at 9.00 and in the afternoon – at 13.00. Table 3 shows that a level of potassium ferrocyanide reduction in the afternoon was lower than in the morning. The decrease of the rate of the Hill reaction may be explained by the increase of starch content in the chloroplasts in the afternoon, which is characteristic of soybean plants. Starch overflow of plastids reduces the level of photochemical processes. It may be due to the deterioration of the mode of illumination of light harvesting complexes, mechanical influence of starch on functional membranes and sorption of proteins on the starch particles. Both enzymes of secondary photosynthesis reactions and ferredoxin, which is a surface protein of thylakoid membranes, can be sorbed on the starch. Moreover, a starch binds cations on its surface, in particular Mg^{2+} cations, which are cofactors of the Hill reaction (Mokronosov, Gavrilenko, 1992).

After the RL irradiation in the evening, the level of specific activity of the Hill reaction did not change in the SD and PhPN lines compared to the control. Irradiation with RL in the middle of the night reduced the reaction level of the SD line relative to control in the morning and afternoon, but did not affect the electron transport in the PhPN line. The decrease of the level of photochemical activity of chloroplasts under RL effect on *Triticum aestivum* L. was previously observed by Mokronosov and co-authors (Mokronosov, Gavrilenko, 1992). In their experiments, it was shown that plants grown under RL had two-fold lower Hill reaction rate compared to plants grown under white light. The authors explained this effect by changes of the biosynthesis of chloroplast electron transport proteins. According to authors, the reason of the decrease of the electron transport intensity after RL irradiation is reducing the activity of the water photolysis system in photosystem II. But we supposed that in our experiments a decrease of the rate of potassium ferricyanide reducing by chloroplasts is the result of interruption of night outflow of assimilates in SD lines, since in the PhPN line RL radiation line did not change the photochemical activity of chloroplasts. Moreover, in the SD line after evening RL irradiation the level of Hill reaction also did not change.

Studying the photochemical activity of chloroplasts, in addition to the reducing potassium ferricyanide, we determined the intensity of reduction of the mineral phosphorus content in the reaction mixture. These data were used to evaluate the speed of photophosphorylation processes. The results in

table 4 indicate that no significant change of the rate of photophosphorylation after evening and night RL irradiation in both lines occurred. The absence of the photophosphorylation decrease is notable as a result of interruption of the night by RL in the SD line, because there was a decrease of the level of potassium ferricyanide reducing. A decrease of the rate of potassium ferricyanide reducing indicates a decrease of the rate of non-cyclic electron transport. If so, then the proton gradient generated by non-cyclic electron transport should also decrease. The latter may be a decrease of the intensity of photophosphorylation. At the same time, literature data indicate that cyclic photophosphorylation can be compensated by reducing the rate of non-cyclic transport. In cyclic photophosphorylation, only the first photosystem is involved and the process is not related to the reducing of potassium ferricyanide (Mokronosov, Gavrilenko, 1992). Thus, the contradictions between a decrease of non-cyclic electron transfer in the SD line as a result of an interruption of night by RL and the unchanged level of photophosphorylation can be explained by the compensation of a decrease of non-cyclic photophosphorylation by an increase of the cyclic photophosphorylation intensity.

Table 3.
Photochemical activity of chloroplasts ($\mu\text{mol K}_3\text{Fe(CN)}_6/\text{mg}$ chlorophyll per hour) of the SD and PhPN isogenic soybean lines under different RL irradiation terms

Variant	Number of exposures	Photochemical activity in the morning (9.00)		Photochemical activity in the afternoon (13.00)	
		Exposure time			
		Before dark period	In the middle of night	Before dark period	In the middle of night
SD line, genotype $E_1E_2E_3$					
Control group	–	281 ± 14	281 ± 14	255 ± 11	255 ± 11
RL irradiation	4	297 ± 15	225* ± 13	270 ± 11	217* ± 15
Control group	–	275 ± 13	275 ± 13	241 ± 10	241 ± 10
RL irradiation	7	289 ± 15	226* ± 10	256 ± 12	206* ± 10
PhPN line, genotype $e_1e_2e_3$					
Control group	-	290 ± 14	290 ± 14	248 ± 12	248 ± 12
RL irradiation	4	285 ± 16	259 ± 13	243 ± 14	236 ± 15
Control group	-	272 ± 14	272 ± 14	239 ± 13	239 ± 13
RL irradiation	7	288 ± 15	253 ± 15	255 ± 14	226 ± 15

* differences with control are significant: $P \leq 0.05$.

Table 4.
Photophosphorylation ($\mu\text{mol KH}_2\text{PO}_4/\text{mg}$ chlorophyll per hour) in isolated chloroplasts of SD and PhPN isogenic soybean lines under different RL irradiation terms

Variant	Photophosphorylation in the morning (9.00)		Photophosphorylation in the afternoon (13.00)	
	Exposure time			
	Before dark period	In the middle of night	Before dark period	In the middle of night
SD line, genotype $E_1E_2E_3$				
Control group	118 ± 5.9	118 ± 5.9	105 ± 6.7	105 ± 6.7
RL irradiation	113 ± 5.5	110 ± 6.3	101 ± 6.6	98 ± 5.3
PhPN line, genotype $e_1e_2e_3$				
Control group	114 ± 6.1	114 ± 6.1	104 ± 4.7	104 ± 4.7
RL irradiation	118 ± 6.7	117 ± 6.6	108 ± 14	109 ± 4.8

* differences with control are significant: $P \leq 0.05$.

The intensity of functioning of the photosynthetic apparatus and the content of chlorophyll, by which we calculated the rate of Hill reaction and photophosphorylation, in some cases did not change in one direction. Therefore, we have studied the effect of RL on indicators that take both of these values into

account at the same time. We analyzed the effect of RL irradiation on the Hill reaction level and photophosphorylation based on the chlorophyll of one leaf (second from the apex) (Table 5). For this purpose, in each experiment we determined the weight of leaves, and knowing the chlorophyll content in one gram of leaves and the photochemical activity attributed to one milligram of chlorophyll, made the appropriate calculations. These indicators are also interesting because they also take into account the size of leaves, which also changed after RL irradiation in the SD line. Data obtained (Table 5) indicate that evening irradiation by RL in the SD line resulted in an increase of ferricyanide reducing and photophosphorylation of one leaf by 38 % and 27 %, respectively, and interruption of night by RL reduced these rates by 45 % and 36 %, respectively, compared to the control. In PhPN plants after irradiation the intensity of ferricyanide reducing and photophosphorylation based on chlorophyll of 1 leaf did not change significantly.

Table 5.
Photochemical activity of chloroplasts of the SD and PhPN isogenic lines under different RL irradiation terms

Variant	Hill reaction ($\mu\text{mol K}_3\text{Fe}(\text{CN})_6/\text{chlorophyll of 1 leaf} \cdot \text{hour}$)		Photophosphorylation ($\mu\text{mol KH}_2\text{PO}_4/\text{chlorophyll of 1 leaf} \cdot \text{hour}$)	
	Exposure time			
	Before dark period	In the middle of night	Before dark period	In the middle of night
SD line, genotype $E_1E_2E_3$				
Control group	110 ± 16	110 ± 16	47 ± 4.9	47 ± 4.9
RL irradiation	152* ± 18	61* ± 14	60* ± 5.5	30* ± 5.2
PhPN line, genotype $e_1e_2e_3$				
Control group	109 ± 15	109 ± 16	46 ± 4.1	46 ± 4.1
RL irradiation	124 ± 17	90 ± 14	51 ± 5.7	42 ± 4.6

* differences with control are significant: $P \leq 0.05$.

Thus, our research showed that photosynthetic apparatus of plants of isolines of the same variety, but with different reactions to the duration of photoperiod, responded differently to red light. It is known that the phytochrome system is important for plants to perceive alternating day and night. "Matching" phytochrome signals with circadian rhythms, plants can respond to the duration of photoperiod (Salomé et al., 2002; Lagercrantz, 2009). It can be assumed that control of the photosynthetic apparatus by the phytochrome system and participation in the photoperiodic reaction of plants are interrelated, because red light induced changes of the chlorophyll content and photochemical activity of chloroplasts in the short-day line. However, in the photoperiodical neutral line changes were not observed. It is likely that a state of the *E* (dominant or recessive) genes indirectly effect on the photosynthetic apparatus.

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Фотохімічна активність хлоропластів ізогенних за генами *E* лінії сої (*Glycine max* (L.) Merr.) за різних термінів опромінення червоним світлом В.Ф. Тимошенко, В.В. Жмурко

У вегетаційних дослідках вивчали вплив різних термінів опромінення рослин червоним світлом (ЧС, 630 нм) на біомасу, площу листя, вміст хлорофілу, рівень реакції Хілла і фотофосфорилування у короткоденної (КД) і фотоперіодично нейтральної (ФПН) ліній сої. Об'єктами досліджень були ізогенні за генами *E* лінії сої (*Glycine max* (L.) Merr.) сорту Clark. Використовували КД лінію (*E1E2E3*) і ФПН лінію (*e1e2e3*). Рослини вирощували у вегетаційній камері в ґрунтовій культурі, ґрунт чорнозем. Рослини КД і ФПН ліній вирощували в 9 посудинах об'ємом три літри, у кожній посудині по 10–12 рослин. Протягом дослідження підтримували постійні умови вирощування: температура 20–24/17–20°C (день/ніч), вологість ґрунту 60–70 % від повної вологоємності ґрунту, інтенсивність освітлення 20 клк, фотоперіод 10 годин. Через 4–5 тижнів вегетації після формування другого справжнього листка рослини кожної лінії в трьох посудинах на початку темного періоду (дослід 1) або в середині ночі (дослід 2) протягом 30 хвилин опромінювали червоним світлом слабкої інтенсивності. Для опромінення використовували світлодіоди, що випромінюють в області 630±10 нм. Інші рослини кожної лінії, що не освітлювали червоним світлом, в трьох посудинах, слугували контролем. Фенологічні спостереження показали, що в контролі на короткому дні КД лінія зацвітала через 43±1,8 доби після появи сходів, а фотоперіодично нейтральна – через 44±2,2 доби. Опромінення ЧС, як перед початком темного періоду доби, так і при перериванні ночі червоним світлом, викликало затримку переходу до цвітіння у КД лінії відповідно на

5±1 і на 7±2,2 діб. У фотоперіодично нейтральної лінії зміни терміну цвітіння в результаті опромінення червоним світлом не встановлено. У короткоденної лінії сої активація фітохромів ЧС перед темним періодом доби викликала зростання біомаси, площі листя, вмісту сумарного хлорофілу, зростання відновлення фериціаніду калію і фотофосфорилування ізольованими хлоропластами у розрахунку на хлорофіл одного листка, тоді як переривання ночі ЧС знижувало ці показники, а також рівень реакції Хилла у розрахунку на 1 мг хлорофілу. Впливу ЧС на вивчені показники у ФПН лінії сої не виявлено.

Ключові слова: соя *Glycine max* (L.) Merr., ізогенні лінії, фітохром, фотохімічна активність хлоропластів, морфогенез.

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**Фотохимическая активность хлоропластов изогенных по генам *E* линий сои (*Glycine max* (L.) Merr.) при различных сроках облучения красным светом
В.Ф. Тимошенко, В.В. Жмурко**

В вегетационных опытах изучали влияние различных сроков облучения растений красным светом (КС, 630 нм) на биомассу, площадь листьев, содержание хлорофилла, уровень реакции Хилла и фотофосфорилирование у короткодневной (КД) и фотопериодически нейтральной (ФПН) линий сои. Объектами исследований были изогенные по генам *E* линии сои (*Glycine max* (L.) Merr.) сорта Clark. Использовали КД линию (*E1E2E3*) и ФПН линию (*e1e2e3*). Растения выращивали в вегетационной камере в почвенной культуре, почва чернозем. Растения КД и ФПН линий выращивали в 9 сосудах объемом три литра. В каждом сосуде росло по 10–12 растений. В течение всего опыта поддерживали постоянные условия выращивания: температура 20–24/17–20°C (день/ночь), влажность почвы 60–70 % от полной влагоемкости почвы, интенсивность освещения 20 клк, фотопериод 10 часов. Через 4–5 недель вегетации после формирования второго настоящего листа растения каждой линии в трех сосудах в начале темного периода (опыт 1) или в середине ночи (опыт 2) в течение 30 минут облучали красным светом слабой интенсивности. Для облучения использовали светодиоды, излучающие в области 630±10 нм. Другие растения каждой линии, которые не освещали красным светом, в трех сосудах, служили контролем. Фенологические наблюдения показали, что в контроле на коротком дне КД линия зацветала через 43±1,8 суток после появления всходов, а фотопериодически нейтральная – через 44±2,2 суток. Облучение КС, как перед началом темного времени суток, так и при прерывании ночи красным светом вызвало задержку перехода к цветению у КД линии соответственно на 5±1 и на 7±2,2 суток. У фотопериодически нейтральной линии изменения срока цветения в результате облучения КС не установлено. У короткодневной линии сои активация фитохромов КС перед темным периодом суток вызвала увеличение биомассы, площади листьев, содержания суммарного хлорофилла, восстановления феррицианида калия и фотофосфорилирования изолированными хлоропластами в расчете на хлорофилл одного листа, тогда как прерывание ночи КС снижало эти показатели, а также уровень реакции Хилла в расчете на 1 мг хлорофилла. Влияния КС на изученные показатели у ФПН линии сои не обнаружено.

Ключевые слова: соя *Glycine max* (L.) Merr., изогенные линии, фитохром, фотохимическая активность хлоропластов, морфогенез.

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