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••• ПРИКЛАДНІ АСПЕКТИ РЕГУЛЯЦІЇ РОСТУ, РОЗВИТКУ ТА ПРОДУКТИВНОСТІ РОСЛИН •••

••• PRACTICAL ASPECTS OF PLANTS GROWTH REGULATION, DEVELOPMENT AND PRODUCTIVITY •••

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Photoperiod-induced changes in total nitrogen and soluble protein content in soybean leaves

I.M. Raievska, A.S. Schogolev

Soybean (*Glycine max* (L.) Merr.) is a short-day plant highly sensitive to photoperiod, with this sensitivity largely regulated by maturity (*E*) genes. These genes influence a wide range of developmental processes, including flowering time, morphological traits, hormone levels, and carbon and nitrogen metabolism. Adaptation to photoperiod involves coordinated changes in morphology, physiology, and biochemistry, ensuring timely transition to reproductive development and optimal crop formation. Field experiments were conducted at the experimental plots of the Department of Physiology and Biochemistry of Plants and Microorganisms of V. N. Karazin Kharkiv National University. This study investigated the effects of photoperiod duration on total nitrogen and soluble protein content in the leaves of near-isogenic soybean lines of the Clark variety, differing in *E1–E4* gene combinations. Short-day lines (Clark variety (*e1E2E3E4e5E7*), line L63-3016 (*e1E2E3e4e5E7*), line L 80-5879 (*E1e2e3E4e5E7*) and photoperiod-insensitive lines (L63-3117 (*e1e2E3E4e5E7*), L71-920 (*e1e2e3E4e5E7*) were grown under natural long-day conditions (16 h) until the V3 stage. Half of the plants were then exposed to short-day conditions (9 h) for 14 days using blackout treatments. Leaf samples were collected at four time points (before the start of the short day effect, during the short day effect - 7 and 14 days, a week after the short photoperiod effect) to assess total nitrogen and soluble protein levels. The results showed that dominant alleles of *E1* and *E2* delayed the transition to the reproductive phase and significantly affected nitrogen and protein accumulation. Specifically, *E1* reduced total nitrogen under both photoperiods, while *E2* increased it under long-day conditions. Both *E1* and *E2* lowered soluble protein content under short-day exposure. No significant effects of *E3* and *E4* were observed. These results demonstrate that the regulation of nitrogen metabolism and protein synthesis in soybean is closely modulated by the interaction between photoperiod and maturity gene expression.

Key words*: plant development, day length, total nitrogen, soluble protein, soybean, E-genes*

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About the authors:
Raievska I.M. – V. N. Karazin Kharkiv National University, Kharkiv, Ukraine, 4, Svobody Maidan, Kharkiv, Ukraine 61022; i.m.rayevska@karazin.ua; https://orcid.org/0000-0003-3656-8178
Schogolev A.S. – V. N. Karazin Kharkiv National University, Kharkiv, Ukraine, 4, Svobody Maidan, Kharkiv, Ukraine 61022; a.s.schogolev@karazin.ua, https://orcid.org/0000-0002-6563-1415

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Introduction

The transition of plants to flowering is regulated by a complex network involving genetic, hormonal, and environmental factors. In *Arabidopsis thaliana*, key genes for photoperiod and vernalization responses include *CONSTANS (CO)* and *FLOWERING LOCUS C (FLC)*, which antagonistically regulate *FLOWERING LOCUS T (FT)* and influence flowering time (Kinmonth-Schultz et al., 2021). In soybean (*Glycine max* (L.) Merr.), flowering is strongly controlled by photoperiod and a set of key loci, known as *E* genes (*E1–E9*), which modulate daylength sensitivity. Among these, *E1* acts as a major floral repressor, encoding a *B3* domain transcription factor that suppresses *GmFT2a* and *GmFT5a*, florigen genes promoting flowering; its expression is enhanced under long-day conditions, delaying flowering (Cao et al., 2017). The *E2* gene, a homolog of *GIGANTEA (GI)* from Arabidopsis, plays a role in the circadian clock and photoperiod response by modulating CO-like genes that influence *FT* expression (Watanabe et al., 2011). *E3* and *E4* encode phytochrome A family photoreceptors (PHYA3 and PHYA2), which detect red/far-red light and contribute to photoperiod sensitivity; mutations in these genes reduce long-day sensitivity, enabling soybean adaptation to higher latitudes. The interactions among these genes shape soybean adaptation to specific latitudes, with *E1* and *E3/E4* mutations enabling early flowering in long-day environments (Li et al., 2018). Moreover, lines carry dominant alleles of *E1* or *E2E3* flower later than those with recessive alleles; notably, *E3* delays flowering in the absence of a dominant *E2* allele but has limited effect under short-day conditions (Raievska et al., 2023).

Among essential nutrients, nitrogen (N) is required in large amounts by soybean plants, and its deficiency can significantly limit growth and development (Staniak et al., 2024). Nitrogen participates in numerous physiological and metabolic processes and is a key structural component of proteins, enzymes, and nucleic acids (Maathuis, 2009). It also forms part of chlorophyll, cytochromes, phytohormones (e.g., cytokinins, auxins), vitamins, and secondary metabolites such as alkaloids, glucosinolates, and cyanogenic glycosides (Leghari et al., 2016). These functions support plant growth, biomass accumulation, and ultimately influence crop quality (Shepilova et al., 2011; Lyashenko et al., 2019; Anas et al., 2020).

Plants require large nitrogen inputs to construct the photosynthetic apparatus, as the rate of photosynthesis is closely linked to tissue nitrogen levels, particularly due to the high demand for the RuBisCo enzyme (Peng et al., 2021). Adequate nitrogen improves adaptive responses to drought, including osmotic adjustments, reactive oxygen species homeostasis, and increased levels of free proline, soluble sugars, and superoxide dismutase activity. When nitrogen is sufficient, antioxidant enzyme activities, including superoxide dismutase and glutathione, are elevated compared to nitrogen-deficient conditions (Tariq et al., 2022; Staniak et al., 2023).

Beyond being a vital macronutrient, nitrogen also acts as a signaling molecule that regulates the timing of flowering. In *Arabidopsis thaliana*, nitrogen deficiency accelerates flowering, whereas both excess and total deprivation can delay it. Transcriptome analyses have identified genes and pathways responsive to nitrogen availability that also modulate flowering time, highlighting potential points of integration between nitrogen signaling and developmental regulation (Brown et al., 2017). Soybean, with its high seed protein content (~40%), has a particularly high nitrogen demand (Staniak et al., 2024). Total plant nitrogen content has been shown to influence seed quality (Piper et al., 1999). Nitrogen can accumulate in vegetative organs (leaves, stems, petioles, pods) and, under deficiency, be remobilized to seeds, inducing leaf senescence and reducing photosynthetic efficiency, ultimately lowering yield (Staniak et al., 2024). Nitrogen scarcity also negatively affects root development and impairs the efficiency of legume-rhizobia symbiosis (Schogolev et al., 2021).

For optimal soybean development, it is crucial that plants receive appropriate nitrogen levels during key growth stages, particularly during the transition from vegetative to reproductive development. Nitrogen application at the early flowering stage has been shown to promote carbohydrate redistribution toward reproductive organs, thereby supporting the transition to flowering (Zhou et al., 2019). However, excessive nitrogen input can suppress this developmental shift, delaying the onset of the reproductive phase. In the model plant *Arabidopsis thaliana*, low nitrogen availability induces earlier flowering, whereas elevated nitrogen levels delay this process (Weber et al., 2018; Lin et al., 2017). Moreover, flowering time regulation in *Arabidopsis thaliana* has been linked to tissue-specific nitrate signaling, particularly at the shoot apical meristem, highlighting the complexity of nitrogen's role in developmental transitions (Olas et al., 2019).

Understanding how soybean growth and development respond to varying photoperiods is essential for advancing global food security. As a photoperiod-sensitive crop, soybean exhibits marked differences in flowering time and maturation based on day length. This sensitivity directly influences its adaptability across latitudes and climatic conditions, ultimately impacting yield stability and productivity. Investigating these responses enables the development and cultivation of soybean varieties tailored to diverse environments, supporting more consistent and increased agricultural output. This research focus directly supports the United Nations Sustainable Development Goal 2: "End hunger, achieve food security and improved nutrition and promote sustainable agriculture" (https://sdgs.un.org/goals/goal2). By improving soybean adaptability through photoperiod research, we contribute to resilient and sustainable farming systems, an essential step toward eradicating hunger worldwide.

Based on this, the aim of the study was to assess how photoperiod duration influences the nitrogen and soluble protein content in the leaves of soybean lines isogenic for *E* genes, and to identify the relationship between these biochemical indicators and the timing of the soybean transition to flowering.

Materials and methods

The research was conducted using soybean lines (*Glycine max* (L.) Merr.) of the Clark variety, isogenic for *E* genes and provided by the National Center for Plant Genetic Resources of Ukraine. The study involved short-day lines – Clark (*e1E2E3E4e5E7*), L63-3016 (*e1E2E3e4e5E7*), and L80-5879

(*E1e2e3E4e5E7*) – and photoperiod-insensitive lines – L63-3117 (*e1e2E3E4e5E7*) and L71-920 (*e1e2e3E4e5E7*); since all lines share the same allelic states at the *E5* and *E7* loci (*e5E7*), only the *E1–E4* genes were used to characterize photoperiodic differences. Field experiments were carried out during the 2018, 2019, and 2021 growing seasons on the experimental plots of the Department of Physiology and Biochemistry of Plants and Microorganisms at V. N. Karazin Kharkiv National University, using 1 m² plots with triplicate replicates for each variant. Plants were initially grown under natural long-day conditions (16 hours), after which half were subjected to short-day treatment (9 hours) by covering with light-permeable material from 5:00 p.m. to 9:00 a.m. for 14 consecutive days.

Phenological observations were recorded at four time points: Day 0 (pre-treatment), Day 7 and Day 14 (during treatment), and Day 21 (7 days post-treatment); at the study's onset, all plants were at the V3 stage, and subsequent phases were marked when at least 50% of plants reached the next stage. Vegetative phases (V3–V7) were tracked by the number of fully developed leaves, while generative phases were defined as R1 (flowering initiation, ≥50% of plants with a flower at any main stem node) and R3 (pod initiation, ≥50% of plants with a pod ≥5 mm on one of the top four main-stem nodes with fully developed leaves).

Total nitrogen and soluble protein were analyzed from the second and third leaves of five randomly selected plants, collected at 8:00 a.m. at each sampling point. Total nitrogen content was determined using the Kjeldahl method (Aguirre, 2023). Soluble protein content was measured using the Lowry method with Miller's modification (Miller, 1959).

Statistical analysis. Statistical evaluation of the data was performed using Statistica 7.0 software (StatSoft Inc., USA). Descriptive statistics, including mean values (x) and standard deviations (SD), were calculated. Intergroup differences were analyzed using Tukey's post hoc test, with statistical significance determined at $P < 0.05$, applying the Bonferroni correction to adjust for multiple comparisons. Mean values were compared separately for each sampling date across all experimental variants — both between long-day (16 h) and short-day (9 h) photoperiods within the same isogenic soybean line, and between different lines under the same photoperiod conditions.

Results and Discussion

Determination of phenological phases of development.

At the initial stage of the study, we assessed the effect of photoperiod duration on the progression of phenological development phases in soybean lines isogenic for *E* genes during the experimental period (Table 1).

Table 1. Influence of photoperiod duration on the progression of phenological development phases in soybean lines isogenic for E genes; field experiments conducted in 2018, 2019, and 2021

Photoperiod, hours	Experimental day			
	0	7	14	21
Short-day lines				
"Clark" – *e1E2E3E4e5E7* (Clark)				
16	V3	V4	V5 – V6	V7
9	V3	V4	V5 – V6	V7 – R1
L63-3016 – *e1E2E3e4e5E7*				
16	V3	V4	V6	V7
9	V3	V4	V5	V6 – R1
L80-5879 – *E1e2e3E4e5E7*				
16	V3	V5	V6	V7
9	V3	V4 – V5	V6	V7 – R1
Photoperiod-insensitive lines				
L63-3117 – *e1e2E3E4e5E7*				
16	V3 – V4	V4 – V5	V6 – R1	R1
9	V3 – V4	V4	V6 – R1	R1
L71-920 – *e1e2e3E4e5E7*				
16	V3 – V4	V5 – R1	R1	R3
9	V3 – V4	V4 – R1	R1	R3

Note: stages are marked: V3-V7 – vegetative phases of 3-7 leaves, R1 – beginning of flowering; R3 – beginning of bean formation.

In short-day lines, the presence of dominant alleles of the *E1* and *E2E3* genes delayed the transition to flowering; these lines initiated flowering only after 21 days of the study, while under long-day conditions, flowering was not observed, and the plants continued to actively accumulate biomass. Consequently, by

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day 21, short-day lines exhibited significant differences in developmental phases, in contrast to photoperiod-insensitive line lines, which had already reached the same developmental stage under both photoperiod conditions. It is well established that recessive alleles of the *e1* and *e2e3* genes promote earlier flowering in soybean (Xia et al., 2012; Xu et al., 2013; Dong et al., 2022). The findings of our study confirm this, as photoperiod-insensitive soybean lines transitioned to the generative phase (R1) earlier than short-day lines. Specifically, the *e1e2e3E4* line initiated flowering by day 7, and the *e1e2E3E4* line by day 14, under both long- and short-day conditions.

Determination of total nitrogen content in soybean leaves.

The next stage of the study focused on assessing the effect of different photoperiod durations on the total nitrogen content in soybean leaves. The results revealed that, at the beginning of the experiment, the lines exhibited differences in nitrogen content (Fig. 1). Short-day lines carrying recessive alleles of the *e1* gene under both long- and short-day conditions demonstrated higher total nitrogen levels compared to the short-day line with the dominant *E1* allele. In contrast, the photoperiod-insensitive lines did not show significant differences in nitrogen content under varying photoperiod conditions (Fig. 1).

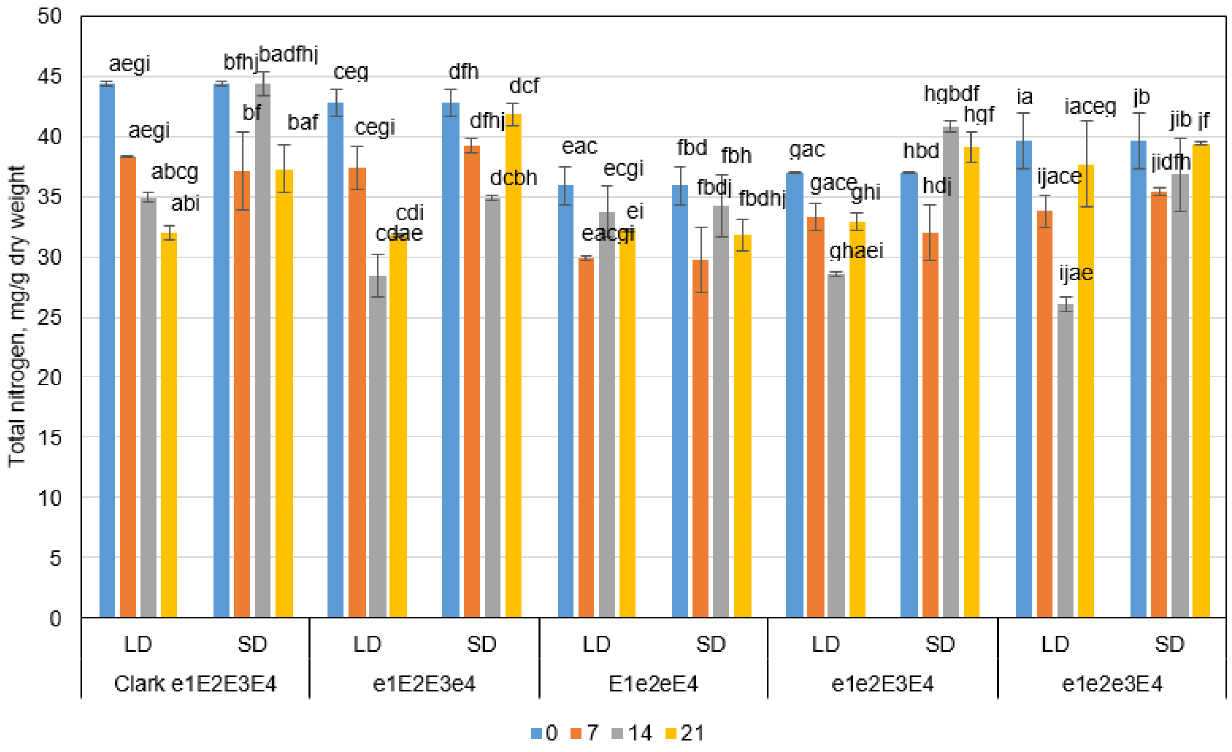

Fig. 1. Effect of photoperiod duration on total nitrogen content in the leaves of soybean lines isogenic for *E* genes. Data represent field experiments conducted in 2018, 2019, and 2021 (n = 15). Note: Mean values were compared separately for each sampling date across all experimental variants — both between long-day (16 h) and short-day (9 h) photoperiods within the same isogenic soybean line, and between different lines under the same photoperiod conditions. Mean values marked with the same letters within a given date differ significantly from each other according to Tukey's test with Bonferroni correction ($P < 0.05$): a – SD line e1E2E3E4 under long (16 h) photoperiod, b – SD line e1E2E3E4 under short (9 h) photoperiod, c – SD line e1E2E3e4 under long (16 h) photoperiod, d – SD line e1E2E3e4 under short (9 h) photoperiod, e – SD line E1e2e3E4 under long (16 h) photoperiod, f – SD line E1e2e3E4 under short (9 h) photoperiod, g – PPI line e1e2E3E4 under long (16 h) photoperiod, h – PPI line e1e2E3E4 under short (9 h) photoperiod, i – PPI line e1e2e3E4 under long (16 h) photoperiod, j – PPI line e1e2e3E4 under short (9 h) photoperiod

On the 7th day of the study, a decrease in the total nitrogen content in the leaves of the studied lines was observed under both long- and short-day conditions; however, the differences in nitrogen content between the lines remained.

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On the 14th day of the study, significant changes were observed in the nitrogen content in the leaves of the short-day line *e1E2E3E4*, specifically a decrease under both long- and short-day conditions. Long-day conditions led to a reduction in leaf nitrogen content in photoperiod-insensitive lines. Under short-day conditions, the lines showed different responses: in the *e1E2E3E4* line, nitrogen content increased, while in the *E1e2e3E4* line, no significant changes were observed compared to day 7.

On the 21st day of the study (7 days after the end of short-day treatment), the short-day lines displayed differences in nitrogen content. In soybean leaves of the Clark variety, nitrogen content decreased; in the *e1E2E3E4* line, it increased; and in the *E1e2e3E4* line, it remained unchanged. In photoperiod-insensitive lines, nitrogen content increased under long-day conditions compared to the 14th day, while under short-day conditions, no significant changes were observed.

Thus, in photoperiod-insensitive soybean lines, nitrogen content in leaves decreased during the transition from vegetative to generative phases under both photoperiod conditions. As reproductive organs developed, nitrogen content increased under long-day conditions but remained stable under short-day conditions.

To determine the influence of maturity genes on total nitrogen content in soybean leaves, we compared lines differing in the allelic composition of the *E1–E4* genes. The results showed that among the gene pairs *E1/e1, E2/e2, E3/e3*, and *E4/e4*, the *E1* and *E2* genes significantly affected nitrogen content. Specifically, the *E1* gene led to a decrease in total nitrogen content under both long- and short-day conditions, while the *E2* gene caused an increase under long-day conditions.

Numerous studies have demonstrated the effects of nitrogen nutrition on the morphological, physiological, and biochemical traits of soybean. In *Arabidopsis thaliana*, nitrogen levels have been shown to influence the timing of the transition from vegetative growth to flowering. This effect may be mediated through the photoperiodic pathway, particularly through the regulation of key flowering genes such as *CONSTANS (CO)* and *FLOWERING LOCUS T (FT)* (Wang et al., 2024). Low nitrogen levels are known to accelerate flowering, while both nitrogen deficiency and excess can delay this transition (Weber et al., 2017; Lin et al., 2017).

However, there is insufficient information on the relationship between nitrogen content in soybean leaves and the transition of plants to flowering. It has been shown that the transition to flowering is regulated by nitrogen and carbon signaling pathways through the modulation of *FLOWERING LOCUS T (FT)* activity in *Arabidopsis thaliana* (Gramma et al., 2024).

Our study demonstrated that both photoperiod length and maturity genes influenced the total nitrogen content in soybean leaves. It should be noted that photoperiod length had a more pronounced effect on this parameter. Photoperiod-insensitive lines, which exhibited faster development and transitioned to flowering earlier under both long- and short-day conditions, showed lower nitrogen content values at the beginning of flowering. However, as they progressed to the pod formation stage, nitrogen content in the leaves increased, likely reflecting the demand for seed development and the activation of nitrogen transport to reproductive organs to meet protein synthesis requirements during the generative phase. In contrast, short-day lines exhibited an opposite response to different photoperiod treatments.

Determination of soluble protein content in soybean leaves.

It is known that approximately 80% of plant proteins are localized in chloroplasts, where they are divided into soluble and insoluble fractions. Around half of the soluble protein fraction consists of ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO). In addition to its role in photosynthesis, the chloroplast also participates in the biosynthesis of key primary and secondary metabolites, including amino acids, fatty acids, and pigments (Kleuter et al., 2024). Photoperiod length influences protein content in leaves. It has been shown that under long-day conditions, *Prunella vulgaris* exhibits increased leaf area, dry weight, and soluble protein content compared to short-day conditions (Li et al., 2022; Xu et al., 2023). In Arabidopsis, long-day conditions lead to elevated levels of enzymes involved in photosynthesis, as well as sucrose and starch biosynthesis (Seaton et al., 2018). According to some authors, this suggests that short photoperiods limit carbon availability, thereby influencing protein metabolism and plant growth (Gibon et al., 2009).

The results of our study showed that, at the beginning of the experiment, the soybean lines did not differ significantly in their soluble protein content (Fig. 2).

After 7 days of exposure to a short-day photoperiod, a decrease in protein content was observed in photoperiod-insensitive lines and plants of the variety, averaging 17%. In photoperiod-insensitive lines, this is likely associated with the transition to the generative phase of development. On the 14th day of short-day exposure, all studied lines exhibited reduced soluble protein levels under short-day conditions. The exception was the photoperiod-insensitive line *e1e2E3E4*, for which the values did not differ from those recorded after 7 days of exposure. Seven days after the cessation of short-day conditions, protein content in leaves did not show significant differences between long- and short-day treatments. Thus, the short-day photoperiod resulted in a reduction in soluble protein content in the leaves of the studied soybean lines compared to the natural long-day condition.

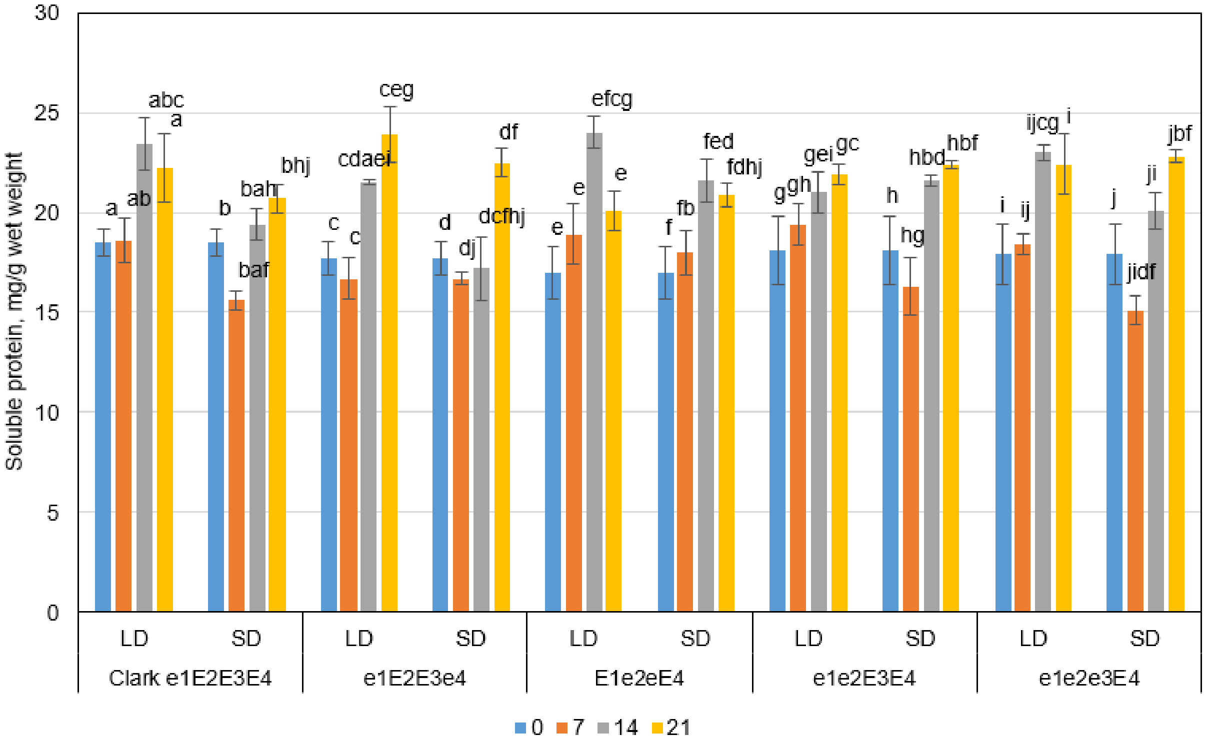

Fig. 2. Effect of photoperiod duration on soluble protein content in leaves of soybean lines isogenic for *E* genes; field experiment, 2018, 2019, 2021 (n = 15).
Note: Mean values were compared separately for each sampling date across all experimental variants — both between long-day (16 h) and short-day (9 h) photoperiods within the same isogenic soybean line, and between different lines under the same photoperiod conditions. Mean values marked with the same letters within a given date differ significantly from each other according to Tukey's test with Bonferroni correction ($P < 0.05$): see Fig. 1 for letters labels description

Analysis of the effect of maturity genes on soluble protein content showed that the dominant alleles of the *E1* and *E2* genes led to decreased protein levels under short-day conditions; however, this effect was observed only after the short-day exposure period.

Thus, soluble protein content in the studied soybean lines changed during the short-day period (7–14 days of the study), indicating a short-term effect, since no significant differences were found at the beginning of the study or after the short-day exposure. Therefore, it can be assumed that among the studied factors, day length is the primary driver influencing changes in protein synthesis.

Conclusion

Thus, the conducted study highlights the primary role of the *E1* and *E2* genes in regulating both the transition of soybean to the generative phase and nitrogen metabolism. The dominant alleles *E1* and *E2E3* delayed flowering compared to lines carrying the recessive alleles of these genes. The *E1* gene consistently exerted a negative effect on total nitrogen content in leaves under all photoperiodic conditions, whereas *E2* contributed to increased nitrogen levels under natural long-day conditions. The *E1* and *E2* genes also reduced the soluble protein content under short-day conditions, indicating their involvement in the photoperiod-dependent regulation of protein metabolism. In contrast, the *E3* and *E4* genes did not exhibit a significant influence on nitrogen and protein metabolism, underscoring the dominant role of *E1* and *E2* in controlling soybean nitrogen balance in response to photoperiod.

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Soluble protein content showed a rapid but reversible response to short-day exposure, suggesting that protein metabolism in soybean leaves is highly dynamic and may be regulated in a more transient manner compared to total nitrogen. The absence of long-term protein differences after return to natural daylengths points to photoperiod-sensitive but developmentally buffered control mechanisms. Although all *E* genes influence photoperiod sensitivity to varying degrees, the lack of significant metabolic changes associated with *E3* and *E4* suggests these genes may act more downstream or in tissue-specific contexts, or that their effects are masked by stronger regulators like *E1* and *E2*. This provides insight into functional differentiation within the *E* gene family.

Understanding the genetic basis of photoperiod sensitivity and nitrogen/protein regulation opens pathways for developing soybean lines with optimized flowering time and nutrient use efficiency. Lines carrying recessive *e1* and *e2* alleles could be valuable for breeding early-flowering, photoperiod-insensitive cultivars suitable for a broader range of latitudes and growing seasons.

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Список використаних джерел / References

Aguirre, J. (2023). The Kjeldahl method. In J. Aguirre (Ed.), *The Kjeldahl method: 140 years* (pp. 53–78). Springer. https://doi.org/10.1007/978-3-031-31458-2_4

Anas, M., Liao, F., Verma, K. K. et al. (2020). Fate of nitrogen in agriculture and environment: Agronomic, eco-physiological and molecular approaches to improve nitrogen use efficiency. *Biological Research, 53*, 47. https://doi.org/10.1186/s40659-020-00312-4

Brown, D., Feeney, M., Ahmadi, M. et al. (2017). Subcellular localization and interactions among rubber particle proteins from *Hevea brasiliensis*. *Journal of Experimental Botany, 68(18)*, 5045–5055. https://doi.org/10.1093/jxb/erx331

Cao, D., Takeshima, R., Zhao, C. et al. (2017). Molecular mechanisms of flowering under long days and stem growth habit in soybean. *Journal of Experimental Botany*, *68(8)*, 1873–1884. https://doi.org/10.1093/jxb/erw394

Dong, L., Hou, Z., Li, H. et al. (2022). Agronomical selection on loss-of-function of *GIGANTEA* simultaneously facilitates soybean salt tolerance and early maturity. *Journal of Integrative Plant Biology*, *64(10)*, 1895–1909. https://doi.org/10.1111/jipb.13332

Gibon, Y., Pyl, E. T., Sulpice, R. et al. (2009). Adjustment of growth, starch turnover, protein content and central metabolism to a decrease of the carbon supply when *Arabidopsis* is grown in very short photoperiods. *Plant, Cell & Environment*, *32(7)*, 859–874. https://doi.org/10.1111/j.1365-3040.2009.01965.x

Gramma, V., Olas, J. J., Zacharaki, V. et al. (2024). Carbon and nitrogen signaling regulate *FLOWERING LOCUS C* and impact flowering time in *Arabidopsis*. *Plant Physiology*, *197(1)*, kiae594. https://doi.org/10.1093/plphys/kiae594

Kinmonth-Schultz, H., Lewandowska-Sabat, A., Imaizumi, T. et al. (2021). Flowering times of wild Arabidopsis accessions from across Norway correlate with expression levels of FT, CO, and FLC genes. *Frontiers in Plant Science*, *12*, 747740. https://doi.org/10.3389/fpls.2021.747740

Kleuter, M., Yu, Y., Verdegaal, L., Pancaldi, F. et al. (2024). Characterizing the extractable proteins from tomato leaves – A proteomics study. *Food Chemistry: X, 25*, 102114. https://doi.org/10.1016/j.fochx.2024.102114

Leghari, S. J., Wahocho, N. A., Laghari, G. M. et al. (2016). Role of nitrogen for plant growth and development: A review. *Advances in Environmental Biology, 10(9)*, 209–219.

Li, Y., Xu, Q., Zhang, Z. et al. (2018). Molecular mechanisms controlling flowering time in soybean. *Journal of Integrative Plant Biology, 60(9)*, 783–798. https://doi.org/10.1111/jipb.13021

Li, Z., Hu, J., Tang, H. et al. (2022). Temperature and photoperiod change the flowering process in Prunella vulgaris by inducing changes in morphology, endogenous hormones, carbon/nitrogen metabolites, and gene expression. *Journal of the American Society for Horticultural Science, 147(2)*, 73–81. https://doi.org/10.21273/JASHS05144-21

Lin, Y.-L., Tsay, Y.-F. (2017). Influence of differing nitrate and nitrogen availability on flowering control in Arabidopsis. *Journal of Experimental Botany, 68(10)*, 2603–2609. https://doi.org/10.1093/jxb/erx053
Maathuis, F. J. M. (2009). Physiological functions of mineral macronutrients. *Current Opinion in Plant Biology, 12*, 250–258. https://doi.org/10.1016/j.pbi.2009.04.003
Miller, G. L. (1959). Protein determination for large numbers of samples. *Analytical Chemistry, 31*(5), 964–966. https://doi.org/10.1021/ac60147a030
Olas, J. J., Van Dingenen, J., Abel, C. et al. (2019). Nitrate acts at the *Arabidopsis thaliana* shoot apical meristem to regulate flowering time. *New Phytologist, 223(2)*, 814–827. https://doi.org/10.1111/nph.15812
Peng, J., Feng, Y., Wang, X. et al. (2021). Effects of nitrogen application rate on the photosynthetic pigment, leaf fluorescence characteristics, and yield of indica hybrid rice and their interrelations. *Scientific Reports, 11*, 7485. https://doi.org/10.1038/s41598-021-86858-z
Piper, E. L., Boote, K. I. (1999). Temperature and cultivar effects on soybean seed oil and protein concentrations. *Journal of the American Oil Chemists' Society, 76(10)*, 1233–1241. https://doi.org/10.1007/s11746-999-0099-y
Raievska, I. M., Schogolev, A. S. (2023). Genetic variation in four maturity genes and photoperiod insensitivity effects on the yield components and on the growth duration periods of soybean. *Regulatory Mechanisms in Biosystems, 14(1)*, 55–63. https://doi.org/10.15421/022309
Schogolev, A. S., Raievska, I. M. (2021). Role of nitrogen deficiency on growth and development near isogenic by E genes lines of soybean co-inoculated with nitrogen-fixing bacteria. *Regulatory Mechanisms in Biosystems, 12(2)*, 326–334. https://doi.org/10.15421/022144
Seaton, D. D., Graf, A., Baerenfaller, K. et al. (2018). Photoperiodic control of the Arabidopsis proteome reveals a translational coincidence mechanism. *Molecular Systems Biology, 14(3)*, e7962. https://doi.org/10.15252/msb.20177962
Staniak, M., Szpunar-Krok, E., Kocira, A. (2023). Responses of soybean to selected abiotic stresses – Photoperiod, temperature, and water. *Agriculture, 13(1)*, 146. https://doi.org/10.3390/agriculture13010146
Staniak, M., Szpunar-Krok, E., Wilczewski, E. et al. (2024). The function of macronutrients in helping soybeans to overcome the negative effects of drought stress. *Agronomy, 14*, 1744. https://doi.org/10.3390/agronomy14081744
Tariq, S., Kumar, M., Alam, B. et al. (2022). Abiotic stress and plant response: Adaptive mechanisms of plants against multiple stresses. In S. Tariq, M. Kumar, B. Alam, S. Kumar, S. Roy, S. Kumar, J. Rane, & S. Kumar (Eds.), *Abiotic Stress in Plants: Mechanisms and Adaptations* (pp. 1–18). Elsevier. https://doi.org/10.1016/B978-0-323-90568-8.00001-8
Wang, M., Wang, J., Wang, Z., Teng, Y. (2024). Nitrate signaling and its role in regulating flowering time in *Arabidopsis thaliana*. *International Journal of Molecular Sciences, 25(10)*, 5310. https://doi.org/10.3390/ijms25105310
Watanabe, S., Xia, Z., Tsubokura, Y. et al. (2011). The soybean FT homolog, GmFT2a, is involved in photoperiodic flowering control. *Plant and Cell Physiology, 52(5)*, 894–903. https://doi.org/10.1093/pcp/pcr042
Weber, K., Burow, M. (2018). Nitrogen – essential macronutrient and signal controlling flowering time. *Physiologia Plantarum, 162(2)*, 251–260. https://doi.org/10.1111/ppl.12664
Xia, Z. J., Watanabe, S., Yamada, T. et al. (2012). Positional cloning and characterization reveal the molecular basis for soybean maturity locus E1 that regulates photoperiodic flowering. *Proceedings of the National Academy of Sciences of the United States of America, 109(32)*, 2155–2164. https://doi.org/10.1073/pnas.1117982109
Xu, M., Xu, Z., Liu, B. et al. (2013). Genetic variation in four maturity genes affects photoperiod insensitivity and PHYA-regulated post-flowering responses of soybean. *BMC Plant Biology, 13(91)*, 1–14. https://doi.org/10.1186/1471-2229-13-91
Xu, Y., Koroma, A. A., Weise, S. E. et al. (2023). Daylength variation affects growth, photosynthesis, leaf metabolism, partitioning, and metabolic fluxes. *Plant Physiology, 194(1)*, 475–490. https://doi.org/10.1093/plphys/kiad507
Zhou, H., Yao, X., Zhao, Q. et al. (2019). Rapid effect of nitrogen supply for soybean at the beginning flowering stage on biomass and sucrose metabolism. *Scientific Reports, 9*, 15530. https://doi.org/10.1038/s41598-019-52043-6
Ляшенко, В. В., Лотиш, І. І., Тараненко, А. О. та ін. (2019). Вплив азотних добрив на урожайність та якість насіння сої. *Scientific Progress & Innovations, (4)*, 58–65. https://doi.org/10.31210/visnyk2019.04.07 [Lyashenko, V. V., Lotysh, I. I., Taranenko, A. O. et al. (2019).

Vplyv azotnykh dobryv na urozhaynist' ta yakist' nasinnya soyi. *Scientific Progress & Innovations*, *(4)*, 58–65. https://doi.org/10.31210/visnyk2019.04.07] (In Ukrainian)
Шепілова, Т. (2011). Вплив добрив та інокуляції насіння на урожайність сої. *Сільськогосподарська мікробіологія, 13*, 117–123. https://doi.org/10.35868/1997-3004.13.117-123 [Shepilova, T. (2011). Vplyv dobryv ta inokulyatsiyi nasinnya na urozhaynist' soyi. *Sil's'kohospodars'ka mikrobiolohiya*, *13*, 117–123. https://doi.org/10.35868/1997-3004.13.117-123] (In Ukrainian)

Вплив тривалості фотоперіоду на вміст загального азоту та розчинного білку у листках сої

І.М. Раєвська, А.С. Щоголєв

Соя (*Glycine max* (L.) Merr.) є культурою короткого дня, яка виявляє високу чутливість до тривалості світлового періоду. Ця чутливість значною мірою визначається генами зрілості (гени *E*), що регулюють широкий спектр процесів у рослин: від строків цвітіння до морфології, гормонального балансу, а також вуглецевого й азотного обміну. Адаптація до змін фотоперіоду супроводжується комплексними морфологічними та фізіолого-біохімічними змінами, які забезпечують своєчасний перехід рослин до цвітіння й формування врожаю. Польові дослідження проводилися на експериментальних ділянках кафедри фізіології і біохімії рослин та мікроорганізмів Харківського національного університету імені В. Н. Каразіна. У межах дослідження вивчали вплив різної тривалості світлового дня на вміст загального азоту та розчинного білка в листках майже ізогенних ліній сої сорту Clark з різними комбінаціями генів *E1–E4*. Короткоденні лінії (Clark (e*1E2E3E4e5E7*), L63-3016 (*e1E2E3e4e5E7*), L 80-5879 (*E1e2e3E4e5E7*)) та фотоперіодично нейтральні лінії (L63-3117 (*e1e2E3E4e5E7*), L71-920 (*e1e2e3E4e5E7*)) вирощували за умов природного довгого дня (16 годин) до фази V3. Після цього частину рослин піддавали впливу короткого дня (9 годин) протягом 14 днів. Відбір зразків здійснювався чотири рази: до початку впливу короткого дня, на 7-й і 14-й день його дії, а також через тиждень після завершення дії коротким фотоперіодом. Результати показали, що домінантні алелі *E1* і *E2* затримували перехід рослин до репродуктивної фази та істотно впливали на вміст азоту і білка. Зокрема, *E1* спричиняв зменшення загального азоту за обох фотоперіодів, у той час як *E2* підвищував його рівень за умов довгого дня. Обидва гени також знижували концентрацію розчинного білка за дії короткого дня. Значного впливу генів *E3* і *E4* виявлено не було. Отже, взаємодія фотоперіоду та експресії генів зрілості тісно пов'язана з регуляцією азотного обміну та синтезу білка у рослинах сої.

Ключові слова: *розвиток рослин, тривалість дня, загальний азот, розчинний білок, соя, E-гени*

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Про авторів:
Раєвська І.М. – Харківський національний університет імені В. Н. Каразіна, майдан Свободи, 4, Харків, 61022, Україна; i.m.rayevska@karazin.ua; https://orcid.org/0000-0003-3656-8178
Щоголєв А.С. – Харківський національний університет імені В. Н. Каразіна, майдан Свободи, 4, Харків, 61022, Україна; a.s.schogolev@karazin.ua; https://orcid.org/0000-0002-6563-1415

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