

THE EFFECT OF CHILLING TEMPERATURE ON GERMINATION AND EARLY GROWTH OF DOMESTIC AND CANADIAN SOYBEAN (*Glycine max* (L.) Merr.) CULTIVARS

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Abstract. Low positive temperature, has an inhibiting effect on growth, development and other physiological processes of cold-sensitive plants which include soybean. An experiment in Petri dishes investigated the effect of temperature: 28/28°C (control), 10/28°C, 28/10°C, and 10/10°C (imbibition/germination), on germination of seeds of 8 soybean cultivars. Another experiment, carried out using pot cultures, investigated the response of 2-week soybean plants of the same cultivars to a 6-day chilling period. The following temperatures were used: 25/20°C (control), 25/0°C, 10/0°C (day/night). Both experiments tested the response of 6 domestic soybean cultivars ('Aldana', 'Jutro', 'Progres', 'Mazowia', 'Nawiko', and 'Augusta') and 2 Canadian cultivars ('OAC Vision', 'Dorothea') to chilling. The obtained results showed that a temperature of 10°C used during germination (28/10°C), and even to a larger extent during imbibition and germination (10/10°C), clearly reduced the speed of germination, percentage of germinated seeds, and radicle length relative to the control, but it increased catalase activity in sprouts. A chilling temperature of 25/0°C and 10/0°C (day/night) significantly increased leaf electrolyte leakage, free proline content and catalase activity relative to the control, but it decreased the photosynthetic rate and total plant leaf area. Seeds and seedlings of cvs. 'Jutro' and 'Nawiko' were generally the least sensitive to chilling, while 'Aldana' and 'Dorothea' were the most sensitive.

Key words: seeds, seedlings, catalase, EL, proline, photosynthetic rate, leaf area

INTRODUCTION

Low positive temperature, has an inhibiting effect on the growth, development and other physiological processes of cold-sensitive plants which include soybean. Huang and Yang [1995] report that soybean seeds generally germinate at a temperature between 10 and 30°C. However, the rate of germination increases with increasing tem-

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perature and reaches its maximum at 30°C. Other authors also report that the optimal temperature for germination and hypocotyl elongation in soybean is around 30°C [Bharati et al. 1983, Tyagi and Tripathi 1983, Posmyk et al. 2001, Liao FangLei et al. 2011]. Bharati et al. [1983] also found that earlier hydration of seeds at higher temperatures accelerated the rate of germination, while at lower temperatures (10°C) it slowed the rate of germination.

Soybean plants are also sensitive to chilling in the juvenile period; temperatures above zero but below 10°C cause damage to soybean plants resulting from temperature-induced physiological and biochemical changes [Wolfe 1991, McKersie and Leshem 1994]. A distinct increase in proline content belongs to the most frequently encountered changes and it has been observed in soybean seedlings by Heerden et al. [2002] and Yadegari et al. [2007], while in other species by Ait Barka and Audran [1997], Dörffling et al. [1997], Chen and Li [2002], Jian et al. [2005], Posmyk and Janas [2007], Apostolova et al. [2008], and Koc et al. [2010].

Chilling stress also causes the leakage of intracellular electrolytes from tissues as a result of the loss of cytoplasmic membrane integrity [Graninetti et al. 1993, Borowski et al. 1997, Bączek-Kwinta and Kościelniak 1999, Bączek-Kwinta et al. 2004, Borowski and Blamowski 2009]. The destructive effect of chilling on the membranes is even greater in light than in the dark [Szalai et al. 1996]. This is undoubtedly associated with the negative effect of stress also on the process of photosynthesis, which has been found both in soybean [Purcell et al. 1987, Caulfield and Bunce 1988, Wang et al. 1997, Heerden et al. 2003a, b] and in other species [Lu-Cun Fu et al. 1994, Starck et al. 2000].

Similarly to other types of environmental stress, chilling induces the production of H₂O₂ and other reactive oxygen species (ROS) in plants. Under such conditions, plants activate the enzymatic system that prevents ROS accumulation. One of the elements of this system is catalase (CAT EC 1.11.1.6). Increased catalase activity under chilling conditions has been observed in germinating soybean seeds by Posmyk et al. [2001], while in seedlings of soybean and other species by Prasad et al. [1994], Zhang et al. [1995], Kang and Saltveit [2002], Lee et al. [2004], and Posmyk et al. [2005]. Increased synthesis of proline and other substances serving as osmoprotectants and antioxidants (ROS) under the influence of chilling requires large energy inputs from plants, which causes the inhibition of growth and development under stress conditions [Borowski et al. 1997, Borowski et al. 1998, Borowski and Blamowski 2009].

Under Polish conditions, soybean seeds are often sown in insufficiently heated soil and after emergence plants are also exposed to short cold periods. This seems to be the main cause of uneven emergence, delayed flowering and seed set and, in effect, low yield. A solution to this situation is to grow cultivars that are less sensitive to chilling stresses and to breed cold-hardy cultivars. The aim of the present study was to evaluate the sensitivity of 6 domestic and 2 Canadian-bred soybean cultivars to chilling stress at the seed germination stage and at the juvenile stage.

MATERIALS AND METHODS

The presented results are means obtained in two separate experiments conducted twice in 2011 and 2012 in a phytotron of the University of Life Sciences in Lublin.

The first experiment was carried out in the period from 11 March to 25 March and from 8 April to 22 April 2011. The test objects were seeds of 6 domestic soybean cultivars ('Aldana', 'Jutro', 'Progres', 'Mazowia', 'Nawiko', and 'Augusta') and 2 Canadian-bred cultivars ('OAC Vision', 'Dorothea'). Before germination, seeds were surface sterilized by immersing them 3 times in 50% ethanol for 1 minute. After every minute in C₂H₅OH, seeds were immersed in distilled water. Following sterilization, seeds were placed in ø 17 cm Petri dishes lined with 2 layers of filter paper moistened with distilled water. The experiment was performed in quadruplicate, with 50 seeds per replicate for each cultivar studied. Imbibition and germination occurred in the dark at a temperature of 28°C and 10°C. The experiments consisted of four experimental series: 1) 28/28°C (imbibition/germination); 2) 10/28°C; 3) 28/10°C; 4) 10/10°C. The imbibition period was 10 hours; however, if germination took place at a temperature different than imbibition, the dishes were transferred to a different phytotron. In the 28/28°C and 10/28°C variants, due to the high rate of germination, the number of germinated seeds was calculated every day and the experiment was terminated after 7 days when all viable seeds had sprouted. In the 28/10°C and 10/10°C variants, on the other hand, the number of germinated seeds was determined every second day and the experiment was terminated after 15 days. After this period, the following traits were determined: the speed of seed germination and percentage of germinated seeds following Maguire [1962] as well as the length of the radicle (embryonic root) based on 10 randomly selected seedlings from each dish. Catalase activity (CAT EC 1.11.1.6) in radicles was also determined in quadruplicate following Aebi [1984] in 0.5 g FW samples.

The second experiment was conducted in the period from 25 March to 29 April and from 4 May to 7 June 2012. Before sowing, seeds were inoculated with soybean inoculant by coating seeds in a bacterial suspension on perlite medium. Next, four seeds of each cultivar were placed in twelve 2 dm³ pots filled with growing medium made of all-purpose potting soil and sand (3:1). The pots were placed in the phytotron at a temperature of 25/20°C and with a photoperiod of 14/10h (day/night) in fluorescent light with an intensity of 200 µmol·m⁻²·s⁻¹ PAR. During the experimental period, the plants were watered with aged tap water, maintaining the moisture content of the growing medium at a level of 70% FWC (field water capacity). After 2 weeks, when the plants had reached the 2–3 true leaf stage, three subseries (with each subseries consisting of 4 pots) were created by random selection for each cultivar, depending on the temperature that the plants were treated with during the period of six consecutive days and nights: 1) 25/20°C control (day/night); 2) 25/0°C; 3) 10/0°C.

Immediately after the end of chilling treatment of the plants, the following was determined in quadruplicate: leaf electrolyte leakage (EL) following the method described by Kościelniak [1993], catalase activity [according to Aebi 1984], and proline content [according to Bates et al. 1973]. Catalase activity in radicles and leaves was expressed in the unit of U·g⁻¹ FW, where $U \cdot g^{-1} FW = (\Delta E / \text{min} \cdot 1 \cdot 2.3 \cdot 10) / (0.036 \cdot 0.1)$: $\Delta E / \text{min}$ – extinction; 1 – response factor; 2.3 – volume of the mixture in the cuvette; 10 – dilution

factor; 0.036 mM – absorption coefficient for peroxidase; 0.1 – volume of the enzyme extract in the cuvette. Photosynthetic rate measurements were made using an LCA-4 portable infrared gas analyzer 10 hours after the end of treatment of the plants with chilling stress. During the measurements, the temperature in the leaf chamber was ca. 27°C, while the PPFD was the same as during plant growth (200 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). After the next week of plant growth at a temperature of 25/20°C, the experiment was terminated and total plant leaf area was determined using a CI-202 laser leaf area meter (CID).

The results of the analysis and instrument measurements presented in this paper are means from the two experiments conducted. They were subjected to analysis of variance for two-way classification, while the significance of differences was determined by Tukey's range test at the $\alpha = 0.05$ level of significance.

RESULTS

The temperature used, both during imbibition and germination, had a significant effect on the speed of seed germination in soybean plants (tab. 1). Seeds germinated quickest at a temperature of 28/28°C, whereas the rate of germination was 1.6 times slower at 28/10°C and twice slower at 10/10°C. Among the cultivars studied, 'Nawiko' and 'Jutro' showed a high rate of the process in question, while the other investigated cultivars showed a significantly lower rate. The Canadian-bred cultivar 'Dorothea' germinated slowest. The effect of temperatures on the percentage of germinated seeds was clearly lower, since at 28/28°C and 10/28°C the studied cultivars germinated at a similar rate. On the other hand, at 28/10°C and 10/10°C seeds showed a slightly lower mean percentage of germination. Regardless of temperatures used, the cultivars 'OAC Vision' and 'Jutro' showed the highest percentage of germination, while this percentage was lower for the other studied cultivars. Cv. 'Aldana' was characterized by the lowest germination capacity (tab. 2).

Table 1. Effect of chilling temperature (10°C) during the period of imbibition or germination as well as in both studied periods on the speed of seed germination in 8 soybean cultivars (plant · day⁻¹)

Temperature °C Imbibition/ Germination	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
28/28 (control)	11.1	13.4	12.2	12.1	13.9	12.5	11.5	11.3	12.2
10/28	10.6	11.6	10.9	11.2	11.5	11.3	11.3	10.7	11.1
28/10	8.9	9.2	6.9	6.5	10.9	7.5	6.4	5.1	7.7
10/10	5.0	6.8	6.8	6.0	6.5	6.1	6.0	4.2	5.9
Mean	8.9	10.2	9.2	9.2	10.7	9.3	8.8	7.8	

LSD_{0.05}: for temperature – 0.50; for cultivars – 0.84; for interaction – 1.98

Table 2. Effect of chilling temperature (10°C) during the period of imbibition or germination as well as in both studied periods on the percentage of germinated seeds of 8 soybean cultivars

Temperature °C Imbibition/ Germination	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
28/28 (control)	83.0	97.8	92.1	85.1	93.0	93.2	97.2	95.0	92.0
10/28	82.8	97.3	91.5	87.0	93.8	86.6	93.5	91.6	90.5
28/10	80.5	91.6	90.5	78.6	93.2	90.0	98.6	83.4	88.3
10/10	71.1	99.0	91.2	80.2	86.0	86.1	97.0	81.3	86.5
Mean	79.3	96.4	91.3	82.7	91.5	89.0	96.6	87.8	

The speed of seed germination was clearly related to radicle length. Seeds of the studied cultivars produced the longest radicles at 28/28°C. A reduction in temperature to 10°C only during the imbibition period did not affect significantly the trait in question. But the application of this temperature during the period of germination, in particular during seed imbibition and germination, resulted in an almost double reduction in radicle length. Cvs. 'Nawiko' and 'Jutro' were characterized by the longest mean radicle length, while the following cultivars: 'Dorothea', 'Aldana', 'OAC Vision', and 'Mazowia', had significantly shorter embryonic roots (tab. 3).

Table 3. Effect of chilling temperature (10°C) during the period of imbibition or germination as well as in both studied periods on radicle length in 8 soybean cultivars (cm · seed⁻¹)

Temperature °C Imbibition/ Germination	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
28/28 (control)	5.1	7.1	6.2	7.4	8.4	7.6	5.6	5.5	6.6
10/28	4.5	7.7	6.1	5.9	8.3	6.7	5.6	4.7	6.2
28/10	3.5	6.7	5.8	3.7	7.1	5.4	3.4	3.2	4.8
10/10	3.5	4.2	3.2	3.6	4.0	4.0	3.2	3.1	3.6
Mean	4.1	6.4	5.3	5.1	6.9	5.9	4.4	4.1	

LSD_{0.05}: for temperature – 0.66; for cultivars – 1.11; for interaction – 2.59

The temperatures used in the present experiments during the period of seed imbibition or germination significantly affected catalase activity in radicles. The activity of this enzyme in radicles that grew at 28/28°C and 10/28°C was nearly twice lower than in radicles produced at 28/10°C and 2.6 times lower than in radicles that grew at

10/10°C. But no significant differences were found in the value of the trait in question between the cultivars analysed, although the activity of this enzyme in cv. 'Jutro' was clearly higher than in cv. 'Dorothea' (tab. 4).

Table 4. Effect of chilling temperature (10°C) during the period of imbibition or germination as well as in both studied periods on catalase activity in radicles of selected soybean cultivars ($U \cdot g^{-1} FW$)

Temperature °C Imbibition/Germination	Cultivars				Mean
	Jutro	Nawiko	Aldana	Dorothea	
28/28 (control)	44.1	45.2	44.9	43.2	44.3
10/28	52.6	47.7	49.0	43.9	48.3
28/10	98.3	91.6	84.2	87.8	90.5
10/10	131.0	126.9	114.6	104.8	119.3
Mean	81.5	77.8	73.2	69.9	

LSD_{0.05}: for temperature – 19.2; for cultivars – n.s.; for interaction – n.s.

Chilling temperatures also had a large effect on the growth of soybean plants and cell plasma membranes stability in the juvenile period. Plants under control conditions (25/20°C) showed the lowest mean value of EL. Treatment of young plants over a period of 6 nights with a temperature of 0°C increased the value of EL by 2.86% relative to the control, while during the day and at night by as much as 6.6%; in both cases, this was a significant effect. Regardless of the period when chilling conditions were applied, cv. 'Jutro' showed the lowest electrolyte leakage, whereas in cvs. 'Mazowia' and 'Progres' EL was significantly higher; there were no significant differences between the other cultivars investigated.

Table 5. Effect of short-term chilling on leaf electrolyte leakage (EL) in plants of 8 soybean cultivars (%)

Temperature °C Day/Night	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
25/20 (control)	13.48	12.67	13.77	13.85	13.59	13.83	13.79	13.68	13.58
25/0	16.04	15.89	17.35	17.05	15.98	16.08	16.48	16.63	16.44
10/0	19.23	18.92	21.96	20.84	19.07	20.12	20.65	20.69	20.18
Mean	16.25	15.83	17.69	17.25	16.21	16.68	16.97	17.00	

LSD_{0.05}: for temperature – 0.68; for cultivars – 1.32; for interaction – 2.95

Plants of the studied soybean cultivars showed low leaf free proline content also at a temperature of 25/20°C. Subjection of the plants to short-term chilling stress at night (25/0°C) increased twice the content of this amino acid, while the application of chilling stress both during the day and at night (10/0°C) increased it as much as 3.8 times. The studied cultivars responded differently to chilling temperatures used. Cv. 'Mazowia' showed the lowest proline content under the experimental conditions, while cvs. 'Jutro' and 'Nawiko' were found to show a significantly higher content relative to the above-mentioned cultivar (tab. 6).

Table 6. Effect of short-term chilling on leaf proline content in plants of 8 soybean cultivars ($\mu\text{g} \cdot \text{g}^{-1} \text{FW}$)

Temperature °C Day/Night	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
25/20 (control)	28.4	34.2	31.3	30.0	35.4	29.3	29.1	28.6	30.8
25/0	69.9	76.2	50.9	42.7	83.2	59.5	58.3	54.2	61.9
10/0	110.6	146.7	115.8	85.3	157.7	108.9	104.2	106.6	117.0
Mean	69.6	85.7	66.0	52.8	92.1	65.9	63.9	63.1	

LSD_{0.05}: for temperature – 11.7; for cultivars – 19.5; for interaction – 44.8

Short-term chilling-induced changes in the plants had a significant effect on leaf photosynthesis. The average level of CO₂ assimilation under control conditions was 5.12 $\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$. Chilling treatment of the plants at night reduced the rate of photosynthesis by 20.3%, while chilling treatment throughout 24 hours by as much as 49.0%. Regardless of temperatures used, cv. 'Mazowia' showed the lowest mean level of photosynthesis, while cvs. 'Nawiko' and 'Jutro' demonstrated significantly higher photosynthetic rates (tab. 7).

Table 7. Effect of short-term chilling on the rate of photosynthesis in plants of 8 soybean cultivars ($\mu\text{mol CO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$)

Temperature °C Day/Night	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
25/20 (control)	4.68	5.92	4.95	4.72	6.02	4.85	4.95	4.87	5.12
25/0	3.89	4.78	3.98	3.63	4.45	3.64	4.03	4.21	4.08
10/0	2.71	3.16	2.85	2.14	3.32	2.28	2.32	2.11	2.61
Mean	3.76	4.62	3.93	3.50	4.60	3.59	3.77	3.73	

LSD_{0.05}: for temperature – 0.35; for cultivars – 0.72; for interaction – 1.40

Table 8. Effect of short-term chilling on catalase activity in leaves of soybean seedlings of selected cultivars ($\text{U} \cdot \text{g}^{-1} \text{FW}$)

Temperature °C Imbibition/Germination	Cultivars				Mean
	Jutro	Nawiko	Aldana	Dorothea	
25/20 (control)	20.4	22.6	21.9	19.8	21.2
25/0	61.6	62.9	54.8	52.3	57.9
10/0	87.6	89.5	73.5	76.8	81.8
Mean	56.5	58.3	50.1	49.6	

LSD_{0.05}: for temperature – 22.3; for cultivars – n.s.; for interaction – n.s.

Similarly to seeds, leaves of the plants responded to chilling conditions by an increase in catalase activity, since the mean activity of this enzyme for four selected cultivars was $21.2 \text{ U} \cdot \text{g}^{-1} \text{FW}$. Short-term treatment of the plants with a temperature of 0°C at night induced an increase in catalase activity up to $57.9 \text{ U} \cdot \text{g}^{-1} \text{FW}$, while treatment of the plants with $10/0^\circ\text{C}$ increased catalase activity nearly four times up to $81 \text{ U} \cdot \text{g}^{-1} \text{FW}$. But no significant differences were found in the value of the trait in question between soybean cultivars (tab. 8).

Table 9. Effect of short-term chilling on total leaf area in plants of 8 soybean cultivars ($\text{dm}^2 \cdot \text{plant}^{-1}$)

Temperature °C Day/Night	Cultivars								Mean
	Aldana	Jutro	Progres	Mazowia	Nawiko	Augusta	OAC Vision	Dorothea	
25/20 (control)	1.60	2.38	1.95	1.85	2.45	1.74	1.88	1.94	1.97
25/0	1.28	1.97	1.57	1.48	2.07	1.39	1.52	1.40	1.58
10/0	0.87	1.55	1.21	1.18	1.59	1.12	1.17	1.11	1.22
Mean	1.25	1.97	1.58	1.50	2.04	1.42	1.52	1.48	

LSD_{0.05}: for temperature – 0.17; for cultivars – 0.37; for interaction – n.s.

Chilling temperatures used also distinctly reduced the growth of young soybean plants (tab. 9). The mean total plant leaf area, as influenced by chilling stress at night, decreased by 19.8% relative to the control, while under the influence of chilling stress throughout 24 hours by 38.1%. Chilling also reduced the increase in leaf area to a different extent in the investigated soybean cultivars. The lowest mean total reduction in leaf area relative to the control was found in cv. 'Nawiko' (25.3%) and cv. 'Jutro' (26.1%), the highest one in cvs. 'Dorothea' (35.6%) and 'Aldana' (33.1), whereas in the other cultivars this reduction ranged from 28.1% to 28.7% (tab. 9).

DISCUSSION

The obtained results showed that soybean seeds, under optimal temperature conditions for their germination (28/28°C) [Bharati et al. 1983, Tyagi and Tripathi 1983, Posmyk et al. 2001, Liao Fang Lei et al. 2011], germinated at a rate of 12.2 plant·day⁻¹ and the percentage of germination was 92% over a period of 7 days. It was similar in the case of seeds in the 10/28°C treatment, but even a 10-hour chilling period during imbibition caused a small decrease in the speed of germination and percentage of germinated seeds. It seems that this can be explained by slower diffusion of water molecules into the interior of seeds due to higher water viscosity. A much larger reduction in the speed of germination at 10°C (28/10°C, 10/10°C), clearly corresponding to radicle length, probably resulted from the decreased activity of numerous enzymes involved in the degradation of seed storage reserves, transport of degradation products, and their metabolism in the embryonic roots. The reason for this was probably the generation of reactive oxygen species (ROS) induced by chilling stress. This can be indicated by the high activity of catalase (CAT) in the radicles of the soybean plants as one of important antioxidants under the conditions of excessive ROS production. Posmyk et al. [2001] also observed an increase in CAT activity in soybean seeds germinating under chilling conditions. It can be presumed that the activity of the antioxidant system was sufficiently high so that under these conditions ROS did not cause significant damage to the embryonic cells but only a slowing of the processes responsible for germination, which seems to be evidenced by the small effect of a temperature of 10/10°C on the percentage of germinated seeds.

The study results showed that short-term chilling significantly induced leaf electrolyte leakage (EL) in the soybean seedlings studied. A similar response to chilling temperatures has also been observed by Graninetti et al. [1993], Borowski et al. [1997], Bączek-Kwinta and Kościelniak [1999], Bączek-Kwinta et al. [2004], Borowski and Blamowski [2009]. Electrolyte leakage from the leaf mesophyll was significantly higher at a temperature of 10/0°C than at 25/0°C. Szalai et al. [1996] also found that chilling stress was more harmful to plants during the day than at night. Electrolyte leakage was undoubtedly caused by ROS generation under the influence of chilling in leaf mesophyll cells. On the other hand, reactive oxygen species induce lipid peroxidation in cell membranes, the loss of their integrity and, as a result of that, also a partial loss of selective permeability. It also seems that the nearly twofold decrease in photosynthesis under the influence of a temperature of 10/0°C could also have resulted from a similar effect of ROS on the cytoplasmic membranes of chloroplasts. As shown by the study on soybean conducted by Heerden et al. [2003a, b], chilling temperature may also significantly limit photosynthesis by reducing stomatal conductance for CO₂. The negative effect of chilling stress on photosynthesis in soybean plants has also been found by Purcell et al. [1987], Caulfield and Bunce [1988], Wang et al. [1997], Heerden et al. [2003a, b], while in other species by Lu-Cun Fu et al. [1994] and Starck et al. [2000].

The obtained results showed that chilling stress significantly increased free proline accumulation in soybean leaves. Other authors also confirm that proline accumulation is an important element of tolerance of soybean [Heerden et al. 2002, Yadegari et al. 2007] and other species [Ait Barka and Audran 1997, Dörffling et al. 1997, Chen and Li

2002, Jian et al. 2005, Posmyk and Janas 2007, Apostolova et al. 2008, Koc et al. 2010] to chilling stress. Chen and Lin [2002] even report that the intracellular level of proline above $2 \mu\text{moli}\cdot\text{g}^{-1}\text{FW}$ in maize plants protected them almost completely against lipid peroxidation. Increased CAT activity, found in the present study, also protects plant cells against lipid peroxidation by H_2O_2 under chilling conditions. A similar response of plants has also been observed by other authors [Prasad et al. 1994, Zhang et al. 1995, Kang and Saltveit 2002, Lee et al. 2004, Posmyk et al. 2005].

The overall effect of chilling stress, in particular long-term stress, on plants is the inhibition of their growth, which has also been observed in cucumber, sweet pepper, and basil in earlier studies of the present author [Borowski et al. 1997, 1998, Borowski and Blamowski 2009]. This results primarily from the inhibition of photosynthesis under these conditions, but also from the activation of energy-expensive adaptive and defensive processes as well as repair processes.

The presented elements of plant responses to chilling temperatures are generally known in the literature. In this study, they were a tool for evaluation of sensitivity of six domestic and two Canadian soybean cultivars to this type of stress. This research shows that the studied soybean cultivars differed in the level of sensitivity to chilling at the stage of seed germination and at the juvenile growth stage. Cvs. 'Jutro' and 'Nawiko' proved to be the least sensitive to chilling; it is probably genetically determined and is associated with the selection towards resistance to this kind of stress.

CONCLUSIONS

A temperature of 10°C used during imbibition or imbibition and germination of soybean seeds clearly reduce the speed of germination, percentage of germinates seeds, and radicle length relative but it increase catalase activity in sprouts.

Seeds of cvs. 'Jutro' and 'Nawiko' are generally the least sensitive to chilling temperature during the germination period, whereas those of 'Aldana' and 'Dorothea' are the most sensitive.

Chilling stress during night ($25/0^\circ\text{C}$), and even to a larger extent during day and night, ($10/0^\circ\text{C}$) induced significant leaf injury (an increase in electrolyte leakage, free proline content and catalase activity, but a decrease in photosynthetic rate and total leaf area) in juvenile soybean plants.

Plants of cvs. 'Jutro' and 'Nawiko' are the least sensitive to chilling temperature at the juvenile stage, while plants of 'Dorothea', 'Mazowia' and 'Aldana' are the most sensitive, so we recommended cvs. 'Jutro' and 'Nawiko' to practice.

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**WPLYW TEMPERATURY CHŁODOWEJ NA KIELKOWANIE
I WCZESNY WZROST KRAJOWYCH I KANADYJSKICH ODMIAN SOI
(*Glycine max* (L.) Merr.)**

Streszczenie. Niska dodatnia temperatura wpływa hamująco na wzrost, rozwój i inne procesy fizjologiczne roślin chłodowrażliwych, do których należy soja. W doświadczeniu prowadzonym na płytkach Petriego badano wpływ temperatury 28/28°C (kontrola), 10/28°C, 28/10°C i 10/10°C (pęcznienie/kielkowanie) na przebieg procesu kielkowania nasion 8 odmian soi. Natomiast w doświadczeniu prowadzonym metodą kultur wazonowych badano reakcję 2-tygodniowych roślin soi tych samych odmian na 6-dniowy okres chłodu. Zastosowano następujące temperatury: 25/20°C (kontrola), 25/0°C, 10/0°C (dzień/noc). W obu doświadczeniach testowano reakcję na chłód 6 krajowych (Aldana, Jutro, Progres, Mazowia, Nawiko, Augusta) i 2 kanadyjskich (OAC Vision, Dorothea) odmian soi. Na podstawie uzyskanych wyników stwierdzono, że temperatura 10°C zastosowana w okresie kielkowania 28/10°C), a szczególnie w okresie pęcznienia i kielkowania (10/10°C), wyraźnie zmniejszyła względem kontroli szybkość kielkowania, procent wykiełkowanych nasion i długość wytworzonego korzenia zarodkowego, a zwiększyła aktywność katalazy w kielkach. Temperatura chłodowa 25/0°C i 10/0°C w istotny sposób zwiększyła względem kontroli wypływ elektrolitów z liści, zawartość wolnej prolina i aktywność katalazy, a obniżyła intensywność fotosyntezy i łączną powierzchnię liści na roślinach. Najmniej wrażliwe na chłód były nasiona i siewki odm. Jutro i Nawiko, a najbardziej wrażliwe Aldany i Dorothea.

Słowa kluczowe: nasiona, siewki, katalaza, EL, prolina, intensywność fotosyntezy, powierzchnia liści

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