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GERMINATION OF OAT SEEDS, CHARACTERIZED BY DECREASED VIGOUR, UNDER DROUGHT CONDITIONS

KIEŁKOWANIE ZIARNIAKÓW OWSA O OBNIŻONYM WIGORZE W WARUNKACH SUSZY

Abstract: In three series of experiments the effects of temperature and drought stress, the drought being simulated with polyethylene glycol (PEG) solution (8000) (–1.5 MPa), on the capacity and dynamics of germination of two oat cultivars – naked Akt and husked Bajka, were determined. The results for imbibition evaluated at 5 °C and 10 °C indicated a greater rate of water uptake in the naked form, under the conditions of both drought and the control. Under the conditions of the control, at varied temperatures of 5, 10, 15 and 20 °C, germinability was found to be significantly lower, by 15 % on average, in the naked form. Under drought conditions no germination only in the case of the naked form (4 %). The dynamics of germination, evaluated under drought conditions at 5 °C and 10 °C, as well as after placing the seeds between layers of blotting paper moistened with water, the temperature being 10, 15 and 20 °C, was significantly higher in naked seeds, which may indicate higher resistance of that oat form to water deficiency in the substrate at the initial phase of germination.

Keywords: naked oat, husked oat, drought stress (PEG), impact of temperature, germinability, dynamics of germination

Germination modifies the potential for growth and development of a plant, and its course is strongly stimulated by external factors and seed characteristics. The start of the imbibition process, essential for activating the enzymes responsible for growth and development of the seed embryo, is conditioned, on the one hand, by the amount of water available from the substrate, and on the other – by the temperature range, most favourable for the course of the germination process [1]. Many plants, including oat, seem to be especially sensitive to water deficiency, during germination in early spring, when a low temperature of soil makes it impossible for them to use fully the over-winter

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soil water. Water stress occurring at that time may delay and decrease germination or totally stop that process [2]. In further stages of development the fibrous root system often suffers from water deficiency when spring frost occurs. Besides, cereal cultivars with husked and naked seeds differ significantly in the course of germination, which results from varied swelling rate [3]. In husked seeds the swelling phase is longer and germination, compared to naked seeds, is delayed. However, naked seeds are characterized by considerable susceptibility to mechanical damage during threshing and ennobling of the sowable material, the result of which is decreased germinability and accelerated ageing [4, 5].

The objective of the study was to determine the modifying effect of temperature and drought stress, induced by polyethylene glycol (PEG 8000), on germination of seeds of various botanical forms of oat, characterized by decreased vigour.

Material and methods

The material for the study included seeds of two oat (*Avena sativa* L.) cultivars: husked Bajka and naked Akt multiplied in observation experiments carried out at the Experimental Station of the Department of Plant Breeding and Seed Science in Prusy, in the year 2005. Harvest was performed when the seed moisture level was 15 % on average, Next, threshing was done, using a laboratory threshing machine, its rotary speed being set at 1.6 m/s. The seeds were stored under warehouse conditions.

Twenty-four months after harvest three series of experiments were carried out, during which drought conditions were simulated with the use of polyethylene glycol solution (PEG 8000), its concentration resulting in the osmotic potential of -1.5 MPa, determined according to Michel's formula [6]. In each experiment seeds were placed in Petri dishes (50 seeds per dish), between layers of blotting paper, and then 16 ml of PEG solution were added. Seed samples sown onto blotting paper moistened with water were the control. The determinations were done with three replications.

In the first experiments seeds were incubated at 5 $^{\circ}$ C and 10 $^{\circ}$ C for 6 days in the case of the control, and for 10 days in the case of seeds germinating under drought conditions. During that period, at 24-h intervals, the moisture content was determined using the drier method, and the start of the germination process, ie the occurrence of radicules above 2 mm long, was established.

In the second experiment seeds germinated under drought conditions at 5 $^{\circ}$ C, 10 $^{\circ}$ C, 15 $^{\circ}$ C and 20 $^{\circ}$ C. On the 10th day seed germinability was assessed according to ISTA methodology [7]. Then, after transferring the dishes with the seeds to 20 $^{\circ}$ C, for successive 20 days observations were done in respect of normally germinating seeds, abnormally germinating seeds, healthy but not germinating seeds, as well as moulding and rotting seeds.

In the third experiment, seeds, under drought conditions, were exposed to temperatures of 5 °C and 10 °C for 5 days, then they were rinsed with water and placed in Petri dishes onto a substrate containing water and kept at temperatures of 10 °C, 15 °C and 20 °C. For 5 successive days the dynamics of germination was evaluated using Maguire's equation [8], the normally germinating seeds being removed from the substrate.

The results of the observation made a basis for carrying out three-factor variance analyses using the independent system. The testing was performed according to the fixed model. For statistical analysis the values expressed in percentage terms were transformed into Bliss's angle values, according to the formula $y = \arcsin\sqrt{x}$. In order to estimate the contribution of the specified sources of variability in the total variability of the investigated characteristics, the components of variance were estimated [9] and their percentage was given. The correlations between the characteristics were evaluated on the basis of the Pearson correlation coefficient or linear regression, or square regression. The values presented in Figures, expressed in percentage terms, were retransformed. For statistical calculations and diagrams Statistica 8.0 was used.

Results and discussion

The results for imbibition, evaluated in the first experiment, show that under the conditions of the control the rate of water uptake and the start of germination depended on temperature and the presence or absence of a seed husk. In the naked cultivar Akt the moisture content increased from 31 % to 43 % at a temperature of 5 °C, and from 34 % to 57 % at 10 °C (Fig. 1). In the husked cultivar Bajka, at a lower temperature the moisture content reached 33–44 %, while at 10 °C it ranged from 34 % to 52 %. The values of the direction components of regression equations show that in the naked form prolonged by 24 h incubation resulted in a faster increase in the moisture content (by



Fig. 1. Moisture content of Akt and Bajka seeds and the number of seeds with the radicule > 2 mm long subjected to imbibition at 5 °C and 10 °C under the conditions of the control (Fz – seed frequency) */** – significant at p = 0.05 and p = 0.01, respectively

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4.3 % on average) compared with the husked form (3.8 %). Under the conditions of the control, first radicules, more than 2 mm long, appeared at 10 °C in the naked cultivar Akt (53 %), on the second day of the experiment, at the moisture content of 38 %, and in the husked cultivar Bajka (4 %) 24 h later, at the moisture content of 45 %. The prolonged, by 24 h, incubation caused a linear increase in the number of seeds with the radicule, its length exceeding 2 mm, which in the naked cultivar was higher at 5 °C (about 22 %). However, in the husked form the curvilinear increase in the number of such seeds was observed only at 10 °C. Thus, it seems that a naked form is better adapted to drought, at least at the beginning of the vegetation period, because water contained in the substrate, even at small amounts, has easier access to a seed and encounters no additional barrier, such as a husk. This is confirmed by the results obtained under the conditions of drought when the seed moisture content in the naked cultivar Akt changed from 25 % to 30 % at 5 °C and from 26 % to 31 % at 10 °C (Fig. 2.). In the husked cultivar, at a lower temperature the seed moisture content ranged from 26 % to 32 %, and at a higher temperature – from 27 % to 34 %, and it was of a rectilinear character. Under drought conditions the rate of water uptake in the naked cultivar, determined by the regression equation, was higher by 49 % on average, at a temperature of 10 °C. In the naked cultivar, in the presence of PEG, germination started on the sixth day (5 %) at a temperature of 10 °C and the moisture content of 29 %. The daily increase in the number of germinating seeds, in the cultivar Akt, was 9.7 %, and on the tenth day it finally reached 44 %. In the husked form single seeds with the radicule more than 2 mm long did not appear until the ninth day, at the temperature of 10 °C and the moisture content of 33 %. Similar results concerning the start of germination were observed by Mos et al [10] when studying the reaction of naked and



Fig. 2. Moisture content of Akt and Bajka seeds and the number of seeds with the radicule > 2 mm long subjected to imbibition at 5 °C and 10 °C under drought conditions (Fz – seed frequency). */** – significant at p = 0.05 and p = 0.01, respectively

husked oat cultivars subjected to the accelerated ageing test under the conditions of drought.

The components of variance calculated for the sources of variability determined in the second experiment showed that the greatest effect on the studied factors was exerted by drought conditions (more than 52 %) (Table 1).

Table 1

Source of variability	Degrees of freedom	Germinability	Percentage of normally germinating seeds after 30 days	Percentage of moulding and rotting seeds
Temperature (A)	3	20.4**	1.3 ns	4.5**
Drought conditions (B)	1	52.8**	85.0**	26.3**
Cultivars (C)	1	0.6**	0.2*	29.6**
Interaction				
$A \times B$	3	20.4**	0.7**	1.5*
$A \times C$	3	0.2*	0.2 ns	0 ns
$\mathbf{B} \times \mathbf{C}$	1	0.6**	4.2**	7.3**
$A \times B \times C$	3	0.2*	0 ns	2.8**
Error	32	4.7	8.3	28.0

Significance of differentiation and the percentage of variance components for germinability, as well as the number of normally germinating, moulding and rotting seeds after 30 days of the experiment

* significant at p < 0.05; ** significant at p < 0.01; ns – insignificant.

The temperature of initial incubation, as well as the combined effect of drought conditions and temperature, had a considerable modifying share in the total variability for germinability (20.4 %). The percentage of moulding and rotting seeds depended most on the cultivar (29.6 %). Seeds stored for 24 month under warehouse conditions were characterized by decreased germinability which at an optimal temperature of 20 °C ranged from 58 % in the naked cultivar Akt to 76 % in the husked Bajka. In the seeds samples subjected to the action of PEG no germination in any of the cultivars was observed. Decreased germinability as a result of drought stress had already been observed by Hosnedl and Honsova [11] in barley, by Michalek and Borowski [2] in soybean, by Yildirim et al [13] in vegetables, by Dhanda et al [14] in wheat, and by Zurek [15] in grasses. The maximum osmotic potential used in those research works was -2.05 MPa. Mos et al [16] found that PEG 8000 concentration resulting in the osmotic potential of -1.5 MPa brings about the greatest differentiation in germination of oat genotypes characterized by high initial germinability. The results obtained in the present research work show that PEG concentration resulting in the osmotic potential of -1.5 MPa inhibited germination in both of the investigated oat forms characterized by decreased germinability. Normally germinating seeds could be noted only under the conditions of the control at a temperature exceeding 5 °C (Fig. 3). Germinability of the cultivar Akt was lower by 16 %, on average, as compared with the cultivar Bajka, irrespective of the temperature used. A rise in temperature of additional 5 degrees resulted in an increase in germinability of about 9 % in the husked form. Thus, the



Fig. 3. Germinability of Akt and Bajka seeds under the conditions of the control, depending on temperature

decreased vigour of seeds of the investigated cultivars became apparent, especially in the naked form, which had already been observed in earlier experiments. The decreased vigour, according to Zurek [15], is an important factor limiting seed germination under drought conditions. In the case of the husked form, under the conditions of the control, transferring the seeds for successive 20 days to a temperature of 20 °C resulted in a significant, 20 % increase in the number of normally germinating seeds (Fig. 4). However, further incubation under drought conditions resulted in the occurrence of such



Fig. 4. The percentage of normally germinating seeds after 30 days, depending on drought conditions



Fig. 5. The percentage of moulding and rotting seeds of the investigated cultivars after 30 days of germination, depending on temperature

seeds only in the naked form (4 %). The difference between the naked form and the husked one was also confirmed by the significantly higher, by 21 % on average, number of moulding and rotting seeds in the naked cultivar. The highest frequency of such seeds (43 %) was noted after initial incubation at a temperature of 10 $^{\circ}$ C (Fig. 5).

The germination dynamics index [8] can be used for the evaluation of many stress factors, including the effect of water deficiency on seed germination (Michalek and Borowski 2002). The variance analysis performed for that index in the third experiment showed a significant effect of incubation temperature under drought conditions and the share of this factor in the total variability reached 30 %. Besides, germination dynamics was modified by the temperature applied after transferring the seeds to the substrate containing water (16.4 %), as well as by differentiation between the cultivars (18.7 %) (Table 2). A higher temperature at the initial stage of incubation affected a rise (of about 14 %) in the rate of germination of the investigated cultivars (Table 3). Transferring the seeds, after five days, from drought conditions to the substrate containing water and keeping them at three different temperatures (10 °C, 15 °C and 20 °C) resulted in significant differences in Maguire's coefficient between the two investigated forms. The greatest value (5.9) was found for the seeds exposed to the temperature recommended for cereal seed germination (20 °C). During the whole incubation period the water deficiency in the substrate, in the case of seeds with decreased germinability, delayed germination dynamics in a different way in both the oat forms. The naked cultivar was characterized by a higher, by 14 %, value of germination dynamics (Table 4). The significance of differentiation of individual effects shows that after transferring the seeds to the substrate containing water and keeping them at different temperatures, greater, by 23 % on average, values of Maguire's coefficient were found for the naked cultivar. The naked cultivar seems to be more resistant to water deficiency in

Table 2

Sources of variability	Degrees of freedom	Germination dynamics according to Maguire
Cultivars (A)	1	18.7*
Temperature during incubation in PEG (B)	1	29.9**
Temperature after incubation in PEG (C)	2	16.4*
Interaction		
$A \times B$	2	8.0 ns
$A \times C$	1	8.6 ns
$\mathbf{B} \times \mathbf{C}$	2	0.0 ns
$A \times B \times C$	2	0.0 ns
Error	24	18.5

Significance of differentiation and the percentage of variance components for germination dynamics calculated for seeds stored at varied temperatures during and after incubation in PEG (-1.5 MPa)

* significant at p < 0.05; ** significant at p < 0.01; ns - insignificant.

Table 3

Germination dynamics depending on the temperature applied during incubation in PEG (-1.5 MPa) and after transferring the seeds to the substrate containing water

Incubation in PEG (days 1–5)				
Temperature [°C]	Germination dynamics according to Maguire	LSD		
5	4.8	0.771		
10	5.6			
After incubation i	n PEG and transferring the seeds to the substrate containing wat	er (days 6–10)		
10	5.0			
15	4.6	0.901		
20	5.9			

Table 4

Germination dynamics of oat cultivars and the differentiation of individual effects after incubation in PEG (-1.5 MPa) and transferring the seeds to the substrate containing water

Cultivar	Germination dynamics according to Maguire					
	Total effect	Individual effects at a temperature of [°C]				
		10	15	20		
Akt	5.5	5.5	5.4	5.8		
Bajka	4.9	4.5	3.9	5.9		
LSD	ns	0.806				

a substrate, occurring at the initial phase of the germination process, and its greater resistance to PEG 8000-induced drought should give it, during that critical period, more time for further stages connected with growth and development of seedlings. This is confirmed by the results obtained by Mos et al [10] showing that tolerance to drought in naked cultivars occurs only under the conditions of shortterm osmotic stress, and its prolongation causes in naked oat seeds a greater decrease in vigour indices, as compared with husked seeds.

Conclusions

1. The study has shown a significant effect of drought stress triggered by polyethylene glycol solution, as well as the temperatures applied, on the rate of water uptake and the start of germination.

2. The seeds of the naked cultivar Akt were characterized by a greater rate of water uptake, under the conditions of both the control and drought, and radicules more than 2 mm long were observed in that form earlier – on the second day and the sixth day respectively.

3. Seeds stored for 24 months under warehouse conditions were characterized by decreased germinability, which at an optimal temperature of 20 $^{\circ}$ C and under the conditions of the control reached 58 % in the naked cultivar Akt and 76 % in the husked cultivar Bajka.

4. Water deficiency in a substrate, in cultivars characterized by decreased germinability, delayed the germination process in a different way in both the oat forms – the husked cultivar and the naked cultivar.

5. Under drought stress lasting for five days, at 10 $^{\circ}$ C and 15 $^{\circ}$ C significantly higher germination dynamics was found in seeds of the naked cultivar Akt, which may be proof of higher resistance of this oat form to water deficiency in a substrate at the initial germination phase.

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Abstrakt: W trzech seriach doświadczeń określono wpływ temperatury i stresu suszy symulowanej roztworem PEG (8000) (–1,5MPa) na zdolność oraz dynamikę kiełkowania dwóch odmian owsa – nagoziarnistej Akt oraz oplewionej Bajka. Ocena imbibicji wykonana dla temperatury 5 i 10 °C wykazała szybsze tempo pobierania wody u formy nagoziarnistej zarówno w warunkach suszy, jak i kontroli. Zdolność kiełkowania w zróżnicowanych temperaturach 5, 10, 15 i 20 °C osiągnęła w warunkach kontroli statystycznie istotnie mniejsze przeciętnie o 15 % wartości u formy nagoziarnistej. W warunkach suszy nie zaobserwowano kiełkowania, a przeniesienie nasion do temperatury 20 °C na kolejne 20 dni przyczyniło się do wystąpienia normlanie kiełkujących nasion tylko u formy nagoziarnistej (4 %). Dynamika kiełkowania oceniona w warunkach suszy dla temperatury 5 i 10 °C, a także po przeniesieniu na podłoże z wodą do temperatury 10, 15 i 20 °C wskazuje na statystycznie istotnie większe wartości u ziarniaków nieoplewionych, co może świadczyć o większej odporności tej formy owsa na niedobór wody w podłożu w początkowej fazie kiełkowania.

Słowa kluczowe: owies nagoziarnisty, owies oplewiony, stres suszy (PEG), wpływ temperatury, zdolność kiełkowania, dynamika kiełkowania