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INFLUENCE OF DIFFERENT STORAGE CONDITIONS ON RYE (*SECALE CEREALE* L.) SEED GERMINATION AND ANTIOXIDANT ACTIVITY

The influence of different storage conditions on rye (*Secale cereale* L.) seed germination and antioxidant activity was investigated. Rye seeds of two lines and hybrid Kharkivianka were stored under model conditions of accelerated aging, at unregulated temperature in the depository, at 4°C, and at minus 20°C for 24 months. Pre-storage treatment involved drying seeds at ≤25°C to 5, 6, and 7 % moisture content. Samples were monitored for the seed germination, seedling length, and antioxidant activity in seeds. The results indicate for seed storage at minus 20°C optimum moisture content 5 %, at 4°C – 6-7 %, at unregulated temperature – 6 %. Rye seed storage under accelerated aging for four months resulted seed germination increasing in comparison with the initial at all moisture content levels. Further storage to 24 months resulted to germination decreasing. There are no clear differences for rye seed germination of one genotype for seed storage with the seed moisture of 5 % and 6 %. Modes storage and seed moisture affecting its germination, have little impact on morphometric parameters of seedlings. This may be due by the selective influence of these factors on seeds. There was no relationship between antioxidant activity, indexes of seed germination and morphometric parameters of seedlings at the investigated seed moisture content levels.

Keywords: *rye, seeds, storage, temperature, moisture, germination, antioxidants, line, hybrid*

INTRODUCTION

Among cereals, rye seeds have the shortest longevity under uncontrolled conditions. The rye seed longevity does not exceed those of wheat and triticale [1, 2], and according to Ewart's classification [3], rye seeds belong to microbotics, meaning loss of viability during three-year storage under natural conditions. Long-term storage causes a seed longevity problem and need for optimization of seed storage conditions. Endogenous biochemical processes occurring during seed storage lead to loss of seed viability [4, 5]. In particular, a sigmoid pattern of reduction in seed germination during storage under uncontrolled conditions is known [6]. Activity of processes occurring in seeds during storage depend on temperature, humidity, light intensity and duration (if any) [5, 7].

Lipid peroxidation (LPO) may occur during seed storage, leading to disruption of membranes and impairing their permeability. It is known that these processes can be limited by free radical oxidation inhibitors - antioxidants (AOs), which create optimal conditions for normal metabolism and functions of cells and tissues [8-11]. During imbibition, antioxidant activity in embryos and endosperm almost triples, while LPO increases almost sevenfold. It is believed that this gradual increase in AO activity is possibly associated with the fact that LPO is suppressed by reserve amounts of AO, which accumulated during seed formation [11]. We know about

biochemical changes, in particular changes of antioxidant contents, in sesame (*Sesamum indicum* L.) seeds during one-year storage [12].

Free radical amounts grow, lutein and soluble protein contents decline, glutenins and gliadins degrade, which was accompanied by a significant increase in proteases activity and drop in gluten elasticity, in durum wheat (*Triticum durum* Desf.) seeds under accelerated storage conditions for 14 days at 40°C and 100 % humidity [13]. Other studies determined that 7-month storage of barley seeds in the depository with low moisture content of $\leq 14\%$ did not lead to a decrease in quality of seeds and was related to slight fluctuations in activities of oxidative and antioxidant processes. Storage of barley seeds with the moisture content of 18 % or $> 20\%$ led to a quick drop in seed germination, accumulation of LPO products and reduction in antioxidant system activity [14]. The author concluded that activities of oxidative and reductive processes were one of physio-biochemical mechanisms that determine seed viability. Increased temperature and grain moisture activate free radical processes and LPO, which reduces the seed germination. Studies show that long-term influence of increased moisture content and temperature leads to a significant accumulation of lipid peroxidation products and loss of seed viability. Catalase activity in barley seeds and seedlings is known to rise after accelerated aging of seeds [15].

Activity of antioxidant system also depends on drying regimens. It was demonstrated that after rapid drying (over silica gel) for 6 days from 39,7 % to 7,5 % the germination of *Mimusops elengi* L. seeds decreased from 100% to 70%, but after gradual drying upon equilibrium moisture content of 85 % the seed germination was 95 %. Studies also show more active mobilization of superoxide dismutase, ascorbate peptidase and catalase after quick seed drying [16].

Negative processes during storage are minimized when seed storage conditions are optimal. Such optimal conditions are developed and improved in genebanks to preserve seed genetic diversity, including rye seeds [17, 28]. Data demonstrate that moisture content of 6-7% is optimal for long-term storage of rye seeds at low positive temperature, which allows storing seeds for about 10 years with germination of $\geq 80\%$. No differences were observed in loss of viability of different accession categories (line, population, variety).

At the same time, the breeding, seed production, experimental work is often a need to store long rye seed with low germination, less than 90% certified (DSTU 2240-93). This need can be caused particular by difficulty of rye reproducing as crosspollinated crop. The information on long-term storage of seeds with low germination is insufficient.

The goal of this study was to determine the optimal storage conditions for rye seeds and opportunities to diagnose the seeds germination by antioxidant activity level.

MATERIAL, METHODS AND RESEARCH CONDITIONS

The test material was rye seeds of lines L 1201 B, L 90691 B and hybrid Kharkivyanka (L 90691A/L 961358B), to assessed the capacity for storage under controlled conditions of rye seed with varying degrees of heterozygosity. Seeds were grown on the experimental farm *Elite* of the Plant Production Institute nd. a. V. Ya. Yuriev of NAAS (Kharkiv region, Eastern Forest-Steppe of Ukraine) in compliance with agronomic requirements in 2013 [19]. The research under storage conditions began 6 months after harvest: seeds were air-dried as per special regimens at $\leq 25^\circ\text{C}$ and relative humidity of 25 % using a Munters dehumidifier (Sweden) to a moisture content of 5-7% and were stored under the experimental conditions of "accelerated aging" at 37°C [20] for 4 and 24 months, in the depository with unregulated storage temperature, or in chambers at 4°C and -18°C. The average unregulated temperature in the depository was 9°C. Temperature in the depository fluctuated from minus 18°C to 25°C. The seed germination was evaluated after 1 month and 24 months of storage for vigor, germination and seedling length. Seeds were sprouted at 20°C in compliance with the recommendations for seed testing [21, 22]. The data were processed by methods of variation statistics [23]. The antioxidant contents were determined using a test system with stable free radical DPPH (1,1-diphenyl-2-picrylhydrazylradical) as per the protocol [24].

RESULTS AND DISCUSSION

In the control option - before the experiment, line L 1201 B seed has germination energy 81 %, germination 82 %; in hybrid Kharkivyanka both indexes equaled 85 %. In contrast, the seed lines L 90691 B characterized by low rates of germination energy – 19 % and germination – 20 % (Table 1).

The line L 1201 B seeds after storage in a sealed container under accelerated ageing for four months don't change germination energy at moisture content 5 % and lowered it for seeds with moisture content 6% and 7%. Germination of seed also significantly increased for all three levels of moisture content. Thus, the stimulatory effect on germination was observed after storage for four months. After 24 months of storage, there was a significant decreasing of germination energy and germination, the most – up to 3 % for germination energy of seeds with moisture content 5 % ($t > 1,98$). The differences of the germination for seeds with different moisture was not observed.

Table 1. Rye seed germination energy and germination after accelerated aging

Accession	Storage duration, months, m.	Germination energy, %			Germination, %		
		at moisture content, %			at moisture content, %		
		5	6	7	5	6	7
L 1201 B	0 ¹	81±3,8	81±3,8	81±3,8	82±3,7	82±3,7	82±3,7
	4	84±3,7	69±4,1	76±4,2	96±2,0	95±1,8	98±1,4
	24	3±3,3	25±8,8	26±10,1	30±8,4	33±9,6	32±10,7
L 90691 B	0	19±7,6	19±7,6	19±7,6	20±7,6	20±7,6	20±7,6
	4	25±3,7	29±3,4	29±3,5	37±4,1	38±3,6	44±3,8
	24	0	0	0	0	0	0
Kharkivianka	0	85±3,1	85±3,1	85±3,1	85±3,1	85±3,1	85±3,1
	4	55±4,5	67±4,7	58±5,1	98±1,2	96±1,9	96±2,1
	24	48±7,4	70±6,8	11±4,7	48±7,4	63±7,1	24±6,4

¹ 0 – germination energy and germination baseline before accelerated aging

The germination increasing under accelerated aging was observed in other experiments with seeds of rare wheat species [25], barley [26], maize [27], peas [28], sunflower [29]. It can be explained by the intensification of metabolic processes under storage conditions, which should be considered as a stress factor. In particular, the accelerated aging experiments with seeds of barley and peas activity increasing of catalase in seeds and seedlings was observed. It is an active response on the content of peroxides increasing, formed under the influence of aging [15]. This metabolic processes activation led to the depletion of physiological stress resistance system, which subsequently led to a significant reduction of germination energy and germination.

The germination energy was decreased at all three levels of moisture content in seeds of hybrid Kharkivyanka after four months storage at the least – 18 % for seeds with 6 % moisture content. Germination increased by 11-13 %. Later, after 24 months of storage, germination energy remained almost at the same level for seeds with moisture content 5 % and 6 % and significantly (till 11 %) decreased for seeds with 7 % moisture content. The germination, like the line L 1201 B, after a certain increase substantially decreased. At least this final decline in germination energy and germination took place at moisture content 6 %, and that should be considered as optimum for storage. The lowest rates were for the both indexes at seed moisture content 7 % ($t > 1,98$).

The line L 90691 B seeds had low initial indexes, after four months of storage. We can see a slight (6-10 %) germination energy increasing and almost a twofold germination increasing. After 24 months of accelerated aging seeds has lost for all three levels of moisture content.

It is noteworthy germination energy and seed germination after 24 months of storage in hybrid Kharkiviyanka at the moisture content level 5 % and 6 % was lower than the indexes for line L 1201 B with similar output indexes. This may testify a higher level of physiological homeostasis in heterozygous genotype compared with homozygous rye. In this respect appropriate further investigation.

Analysis of the antioxidant activity (AA) showed that most of the options it was at the same level. A similar trend described for other species [30]. The highest level of AA (47 %) was observed for the seed lines L 1201 B with a moisture content of 5 % (Fig. 1). This option is different from most other sharp by the decline in seeds germination energy: from 81 % to 3 %. We can assume that the AA increasing was a demonstration of mobilization of physiological mechanisms to counteract destructive oxidation processes.

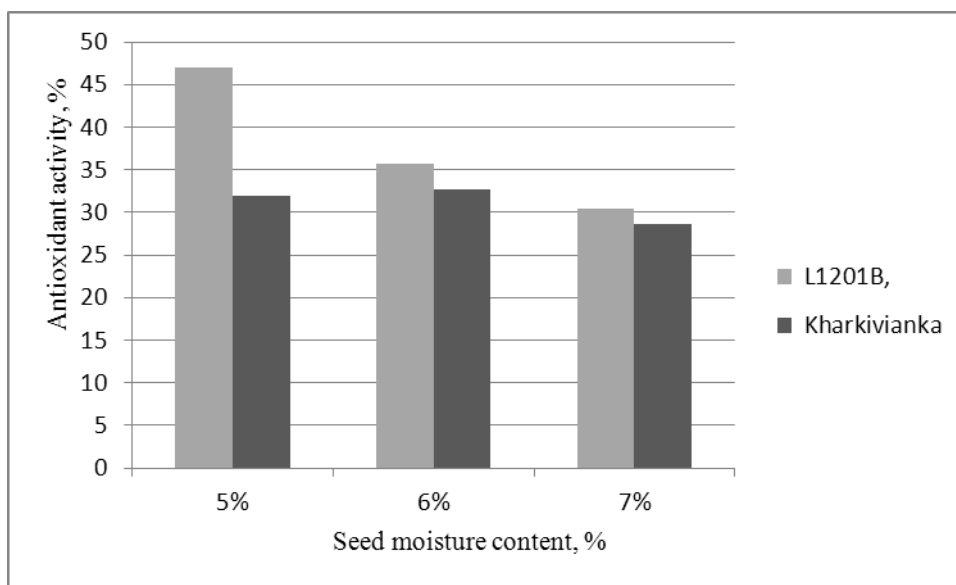


Fig.1. Antioxidant activity in rye seeds with various moisture content, which were stored for 24 months under accelerated aging

To search for conditions of seed storage with lower energy costs, we analyzed the seeds germination with moisture content of 5, 6 and 7 % after 24-month storage in the depository with unregulated temperature as well as in chambers at 4°C and minus 20°C (Table 2).

According to the indexes of germination energy and germination after 24 months of storage, optimum for lines L 1201 B seeds at unregulated temperature conditions were moisture levels 5 and 6 %, at a temperature 4°C – 6 and 7 %, at a temperature 20°C – 5 %.

For hybrid Kharkivianka seeds with the similar germination have the best indexes for seeds with moisture content 6 % at unregulated temperature, 5 % – at a temperature minus 20°C. The germination energy at all three levels of moisture content differ slightly at the temperature 4 °C. The germination was better in the versions with seed moisture content 7 %.

A significant germination energy and seed germination increasing for L 90691 B line was observed for seed with 7 %. The germination is increased, to a lesser extent, also at seed moisture content 5 %. Temperature 4°C was more favorable for the seeds with a moisture content 6 %. The moisture content level doesn't significantly affected on both indexes at minus 20 °C but the trend towards higher germination was observed at 7 % moisture content.

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Table 2. Germination energy and germination of rye seeds accessions after 24 - month storage under different controlled conditions, %

Accession	Storage conditions	germination energy of seeds with moisture content, %			germination of seeds with moisture content, %		
		5	6	7	5	6	7
L 1201 B	0 ¹	81±3,8	81±3,8	81±3,8	82±3,7	82±3,7	82±3,7
	UT ²	68±4,8	59±7,4	46±5,5	86±3,5	89±4,8	73±4,8
	4°C	48±5,9	59±4,7	58±4,8	76±5,1	83±3,6	82±3,7
	-20°C	70±6,1	47±5,5	65±7,0	93±3,4	81±4,3	59±7,3
L 90691 B	0	19±3,3	19±3,3	19±3,3	20±3,4	20±3,4	20±3,4
	UT	21±5,7	13±4,2	47±7,3	37±6,7	24±5,4	60±7,2
	4°C	5±3,4	33±5,8	24±6,1	15±5,5	45±6,1	37±6,9
	-20°C	17±5,2	20±4,8	18±4,7	26±6,1	26±5,3	33±5,8
Kharkivianka	0	85±3,1	85±3,1	85±3,1	85±3,1	85±3,1	85±3,1
	UT	41±8,1	74±6,7	40±7,1	41±8,1	86±5,3	69±6,7
	4°C	66±5,8	67±5,7	56±7,4	73±5,4	81±6,4	89±4,7
	-20°C	81±9,8	57±7,6	63±6,4	94±6,1	69±7,1	70±6,1

¹ 0 – germination energy and germination baseline before accelerated aging;

² UC – depository with unregulated temperature

For hybrid Kharkivianka seeds with the similar germination have the best indexes for seeds with moisture content 6 % at unregulated temperature, 5 % – at a temperature minus 20°C. The germination energy at all three levels of moisture content differ slightly at the temperature 4 °C. The germination was better in the versions with seed moisture content 7 %.

A significant germination energy and seed germination increasing for L 90691 B line was observed for seed with 7 %. The germination is increased, to a lesser extent, also at seed moisture content 5 %. Temperature 4°C was more favorable for the seeds with a moisture content 6 %. The moisture content level doesn't significantly affected on both indexes at minus 20 °C but the trend towards higher germination was observed at 7 % moisture content.

Germination energy of L 1201 B seeds with moisture 5, 6, 7 % storage at 4 °C for 24 months was lower than the initial by 33, 22 and 23% ($t > 1,98$), and the germination is almost no different the initial (see. Table. 2). Germination energy and germination were not found significant differences for seed within specified moisture under this storage mode.

After L 90691 B seeds storage at a temperature 4 °C for 24 months observed decrease in germination energy by 14 % for seeds with a moisture content 5 %, an increase by 14 % at moisture content 6 % ($t > 1,98$) and the absence of significant changes for seed moisture 7 % (see. tab. 2). There was no germination changes for seed with moisture content 5 % ($t < 1,98$), germination increasing by 25 % for seeds with a moisture content 6 % and by 17 % and moisture content 7 % ($t > 1,98$).

Kharkivianka hybrid seeds during storage at low positive temperatures 4 °C during the same period lower rates of germination energy by 20-30 % ($t > 1,98$) observed, but germination is

not different from the initial except the seeds with a moisture content of 5 % where it was lower than the original by 12 %.

Thus, rye seed accessions storage with moisture content 5-7% at 4 °C for 24 months led to reduce the germination energy and had no effect on germination in most cases. Only in rare cases, mostly in the variant with seed moisture content 5 % the storage mode led to its decline.

Germination energy of seeds with moisture 5, 6, 7 % of the sample L 1201 B, stored at a temperature minus 20 °C for 24 months, was below the initial respectively 11, 34 and 16 % ($t > 1,98$). The similarity of this accession is not different from the original version except where seed moisture was 5 % and the germination after the storage term was by 9 % above the initial ($t > 1,98$). A similar lack of differences were for germination in variants with moisture content 5 and 6 %.

Seeds storage of Kharkivianka hybrid at the temperature of minus 20 °C for 24 months led to germination energy decreasing approximately by 25 % at seed moisture 6 and 7 %. Germination energy remained unchanged for seeds with 5 % moisture content. A similar trend was observed for seed germination. Germination decreasing approximately by 15 % ($t > 1,98$), for seed of moisture content 6 and 7 % was observed, and for seeds with 5 % moisture content also remained unchanged. These data suggest the benefit of rye seeds storage at a temperature minus 20 °C at 5 % moisture content.

Antioxidant activity of rye seeds that were stored under different conditions slightly varied (Figure 2). AO activity differed depending rather on genotype than on storage conditions. We believe that such minimal variations were caused by relatively low moisture content in seeds, because it was reported that no monosemous decline in AO activity occurred during long-term storage of seeds with moisture content $\leq 14\%$ [14].

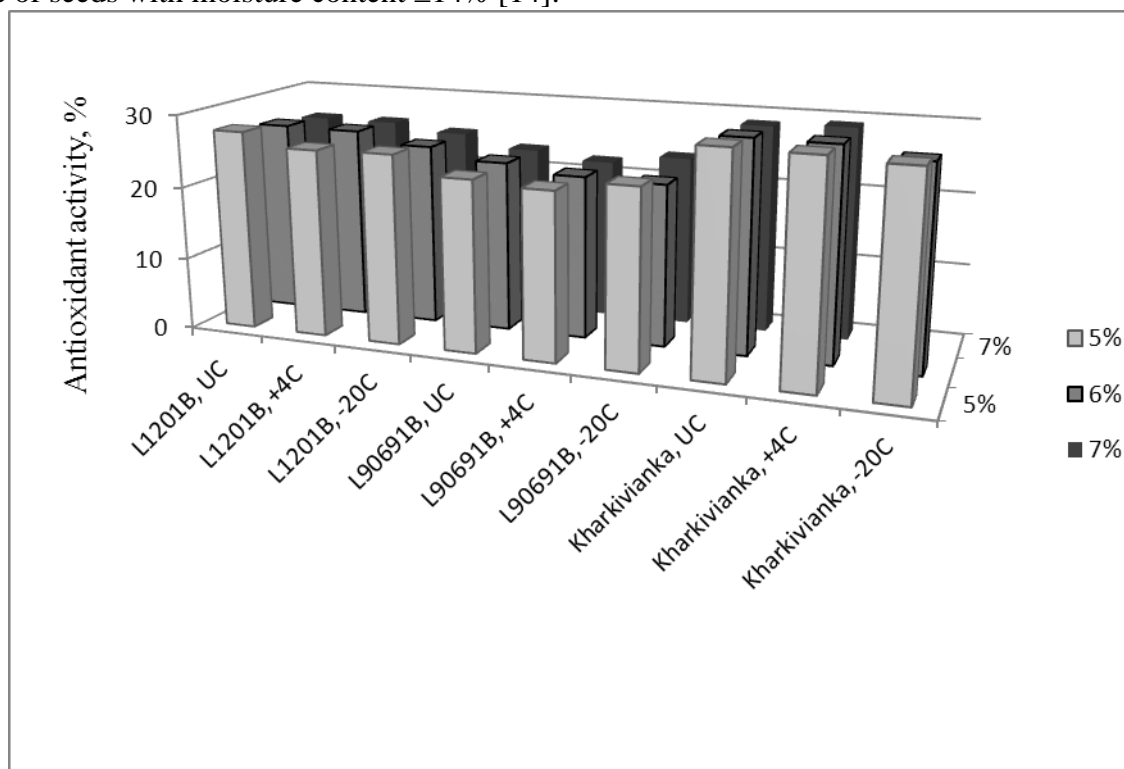


Fig.2. Antioxidant activity of rye seeds after storage under different conditions

The length of seedlings from seeds after different regimes of seed storage was estimated. The length of rye seedlings from seeds that were stored for 24 months did clearly depend either on storage conditions or on moisture level (Table 3). Only accelerated aging regimes caused expected inhibition of shoot growth by 2-5 cm. Morphometric indexes of coleoptiles with leaves and roots were reduced in seedlings from 'Kharkivianka' seeds with 7 % moisture content that were stored at minus 20 °C, but not at unregulated temperature. This can be explained by selective influence on

seed storage modes: in each case germination was less than 100 %, and stronger embryos survived, and relised their potential of vigour growth at a similar level. At the same time, for line L 1201 B seedlings were more longer coleoptile with leaves and roots for seeds storage at unregulated temperature, low positive temperature in comparison with seeds of all three levels of seed moisture content.

Table 3. Length of rye seedlings from seeds stored under different conditions, cm

	Storage conditions	Length (cm) at moisture content, %					
		5		6		7	
		Coleoptile with leaves	Root length	Coleoptile with leaves	Root length	Coleoptile with leaves	Root length
L 1201 B	accelerated aging	5,2±0,9	8,0±0,7	7,6±1,4	6,6±0,7	7,6±2,5	6,6±1,3
	UT ¹	12,6±0,7	10,6±0,4	11,4±1,0	10,0±0,9	12,6±0,9	11,1±0,5
	4°C	11,0±1,0	10,4±0,7	11,7±0,9	11,3±0,7	10,7±0,9	11,2±0,7
	-20°C	9,7±1,4	9,9±1,0	6,8±1,0	10,1±1,1	9,9±1,7	8,4±1,6
L 90691 B	accelerated aging	-	-	-	-	-	-
	UT	10,3±0,7	9,3±,9	9,9±1,3	8,2±0,7	9,3±1,2	9,3±0,6
	4°C	11,8±1,4	9±1,4	11,7±0,7	9,6±0,6	9,0±1,0	9,6±1,0
	-20°C	11,4±1,7	9,2±0,9	9,7±1,3	8,5±1,1	11,2±1,1	9,5±0,5
Kharkivian ka	accelerated aging	5,1±0,9	5,9±0,9	5,6±0,8	7,6±0,9	4,1±1,0	4,5±0,7
	UT	10,6±1,1	9,8±1,1	9,8±1,3	10,2±0,9	7,8±1,5	9,2±0,9
	4°C	10,4±0,9	9,6±0,6	8,3±0,8	9,7±0,6	10,9±0,8	10,2±0,4
	-20°C	10,7±1,0	11,4±0,9	11,7±0,5	10,9±0,3	8,7±1,0	8,3±0,8

¹ UC – depository with unregulated temperature

There was no correlation between morphometric indexes of seedlings and AO activity.

CONCLUSIONS

Our studies show optimal moisture content 5 % for rye seed accessions storage at minus 20 °C, 6-7 % – at 4 °C storage temperature, 6 % – at unregulated storage conditions.

Rye seed storage under accelerated aging for four months resulted an increasing seed germination in comparison with the initial at all seed moisture levels. Further storage up to 24 months resulted to lower germination than the initial. There is no differences for rye seed germination for seed storage at moisture content 5 % and 6 %.

Storage modes and seed moisture content, affecting on germination little impact on morphometric seedling indexes that may be due by selective influence of these factors on seeds.

There were no relationship between seed antioxidant activity, seed germination, and seedlings morphometric indexes at the investigated seed moisture content levels.

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ВПЛИВ РІЗНИХ РЕЖИМІВ ЗБЕРІГАННЯ НАСІННЯ ЖИТА (*SECALE CEREALE* L.) НА ЙОГО СХОЖІСТЬ І АКТИВНІСТЬ АНТИОКСИДАНТІВ

Мета. Визначення оптимальних режимів зберігання насіння жита і можливості діагностувати схожості насіння за рівнем його антиоксидантної активності.

Результати і обговорення. Проведено дослідження впливу різних режимів зберігання насіння жита (*Secale cereale* L.) на його схожість і активність антиоксидантів. Насіння жита двох ліній та гібриду Харків'янка зберігалось в умовах прискореного старіння, сховищах з нерегульованою температурою, температурою 4 °С, температурою мінус 20 °С протягом 24 місяців. Перед закладанням на зберігання насіння висушувалося за температури не вище 25 °С до рівнів вологості 5, 6 і 7 %. Контроль стану насіння проводився за показниками енергії проростання та схожості насіння, довжиною проростків, а також антиоксидантною активністю насіння. Результати досліджень свідчать, що для зберігання насіння досліджених зразків жита за температури мінус 20 °С оптимальною вологістю є 5 %, за температури 4 °С – 6-7 %, за нерегульованих умов – 6 %. Зберігання у режимі прискореного старіння протягом чотирьох місяців мало наслідком підвищення схожості насіння жита проти вихідної за всіх рівнів вологості. Подальше зберігання в умовах прискореного старіння до 24 місяців призвело до зниження схожості порівняно з вихідною. Не встановлено відмін за схожістю насіння жита при зберіганні насіння в умовах прискореного старіння з вологістю 5 % і 6 %. Використані режими зберігання насіння з відповідною вологістю впливають на його схожість, але не відзначено значного впливу на морфометричні показники проростків, що може бути обумовлено селективною дією цих чинників на насінини. Між показниками активності антиоксидантів насіння, його схожості і морфометричними показниками проростків залежності не виявлено.

Висновки. Для зберігання насіння досліджених зразків жита за температури мінус 20 °С оптимальною вологістю є 5 %, за температури 4 °С – 6-7 %, за нерегульованих умов – 6 %. Зберігання у режимі прискореного старіння за температури 37 °С спочатку мало наслідком підвищення схожості насіння за всіх рівнів вологості, в подальшому – зниження схожості порівняно з вихідною. Не виявлено залежності між показниками активності антиоксидантів насіння та показниками життєздатності і морфометричними показниками проростків за досліджених рівнів вологості насіння.

Ключові слова: жито, насіння, зберігання, вологість, температура, схожість, антиоксиданти, лінія, гібрид

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ВЛИЯНИЕ РАЗЛИЧНЫХ РЕЖИМОВ ХРАНЕНИЯ СЕМЯН РЖИ (*SECALE CEREALE* L.) НА ЕГО ВСХОЖЕСТЬ И АКТИВНОСТЬ АНТИОКСИДАНТОВ

Цель. Определение оптимальных режимов хранения семян ржи и возможности диагностировать всхожесть семян по уровню их антиоксидантной активности.

Результаты и обсуждение. Проведено исследование влияния различных режимов хранения семян ржи (*Secale cereale* L.) на его всхожесть и активность антиоксидантов. Семена ржи двух линий и гибрида Харьковчанка хранились в условиях ускоренного старения, хранилищах с нерегулируемой температурой, положительной температурой 4 °С, отрицательной температурой минус 20 °С в течение 24 месяцев. Перед закладкой на хранение семена высушивались при температуре не выше 25 °С до уровней влажности 5, 6 и 7 %. Контроль состояния семян проводился по показателям энергии прорастания, всхожести семян, длине проростков, а также антиоксидантной активности семян. Результаты исследований свидетельствуют, что для хранения семян исследованных образцов ржи при отрицательной температуре минус 20 °С оптимальной влажностью

является 5 %, при температуре 4 °С – 6-7 %, при нерегулируемых условиях – 6 %. Хранение при режиме ускоренного старения в течение четырех месяцев привело к повышению всхожести семян ржи по сравнению с исходной для семян всех уровней влажности. Дальнейшее хранение до 24 месяцев привело к снижению всхожести семян по сравнению с исходной. Не установлено различий по всхожести семян ржи при хранении семян с влажностью 5% и 6% при этом режиме. Режимы хранения и влажность семян, воздействуя на их всхожесть, мало влияют на морфометрические показатели проростков, что может быть обусловлено селективным действием этих факторов на семена. Между показателями активности антиоксидантов семян, показателями всхожести и морфометрическими показателями проростков зависимости не выявлено.

Выводы. Для хранения семян исследованных образцов ржи при температуре минус 20 °С оптимальной влажностью считается 5 %, при температуре 4 °С – 6-7 %, при нерегулируемых условиях – 6 %. Хранение семян в условиях ускоренного старения старения при температуре 37 °С сначала вызывает повышение всхожести семян при всех уровнях влажности, а в дальнейшем – снижение. Не выявлено зависимости между показателями активности антиоксидантов семян, показателями всхожести и морфометрическими показателями проростков при исследованных уровнях влажности семян.

Ключевые слова: *рожь, семена, хранение, влажность, температура, всхожесть, антиоксиданты, линия, гибрид*