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Enrichment of the winter triticale gene pool under intergeneric hybridization

I. Diordiieva[1,*], V. Kochmarskyi[2], Ia. Riabovol[3], L. Riabovol[1] and O. Serzhyk[1]

[1]Uman National University of Horticulture, Uman, Ukraine, Faculty of Agronomy, Department of Genetics, Plant Breeding and Biotechnology, 1 Institytska Str., UA20300 Uman, Ukraine
[2]State Enterprise "Experimental Farm "Elite" of Mironovskyi Institute of Wheat named after V.N. Remeslo NAAS of Ukraine, v. Central, UA08853 Mironovskyi region, Ukraine
[3]Uman National University of Horticulture, Uman, Ukraine, Faculty of Agronomy, Department of Plant Growing, 1 Institytska Str., UA20300 Uman, Ukraine
[*]Correspondence: diordieva201443@gmail.com

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Abstract. The purpose of the research was to expand the genetic diversity of winter triticale under intraspecific and remote hybridization and to obtain new valuable forms for their involvement in the breeding process of creating high-yielding crop varieties. For this purpose interspecific and remote hybridization of different species of wheat, rye, triticale, *Elimus arenarius* L. and evaluation of obtained hybrids were carried out from 2013 to 2020. According to the results of our research it is proved that the species *Triticum spelta* L. and *Triticum petropavlovskyi* Udacz. had dominant genes of incompatibility with rye. Crossing *Triticum compactum* Host. and *Triticum sphaerococcum* Perciv. species with rye allowed obtaining a higher level of seed setting, but the grain obtained with the participation of *Triticum sphaerococcum* Perciv. species was not viable. Compatibility of triticale with spelt and *Elimus arenarius* L. was low. The level of seed formation in F_1 hybrids under artificial pollination was higher than under spontaneous pollination. Seed germination obtained from pollination of F_1 hybrids by fertile forms of triticale - was low. It was found that hybridization of three-species triticale with spelt had a positive effect on grain quality indicators in the offspring. Crossing triticale with *Elimus arenarius* L. led to ear elongation, but caused a significant reduction in all indicators of grain quality in the offspring. As a result of remote hybridization of three-species triticale and spelt wheat, winter triticale varieties Navarra and Strateh were created and included in the State register of plant varieties suitable for distribution in Ukraine since 2018.

Key words: initial material, hexaploid wheat, rye, crosses, varieties.

INTRODUCTION

Triticale is an artificially created biological species that does not have a natural center of origin and a long process of evolution (Lonbani & Arzani, 2011). Therefore, a necessary condition for successful breeding work is the constant production of new initial material involving a wide variety of available forms and remote species, in particular, wheat and rye with the best characteristics in terms of economic-and-valuable features and properties (Gorianina, 2015; Kang et al., 2016).

Synthesis of new materials is possible using different methods, in particular: 1) synthesis of primary octaploid and hexaploid triticale for wheat and rye hybrids and subsequent doubling of the number of chromosomes under the action of polyploidizing substances. This is the main method of introducing wheat and rye germplasm into the triticale gene pool (Sechniak & Sulyma, 1984; Lule et al., 2014); 2) synthesis of secondary hexaploid triticale, based on pollination of F_1 hybrids of different origins by pollen of hexaploid triticale. This method is technically simple and allows creating new forms of crop on a large scale on the basis of highly productive local, selection and foreign samples of wheat, rye and triticale (Sisodia & McGinnis, 1970). The basis of both methods is obtaining of remote hybrids between wheat and rye. Since the genetic diversity of parental forms is great, the number of theoretically possible combinations can be significant (Sechniak & Sulyma, 1984; Sisodia & McGinnis, 1970).

The lack of study of the problem of wheat and rye hybridization and the diversity of scientists' opinions in this regard confirms the relevance, theoretical and applied significance of the scientific field.

The purpose of the research was to expand the genetic diversity of winter triticale samples under intraspecific and remote hybridization and to obtain new valuable forms for their involvement in the selection process of creating high-yielding crop varieties.

MATERIALS AND METHODS

The research was conducted during 2013–2020 at the plots of land of the Department of genetics, plant selection and biotechnology of Uman National University of Horticulture located in the Right-Bank Forest-Steppe zone of Ukraine. The region belongs to the subzone of unstable humidity (there are droughts once in 2–3 years). The average long-term precipitation rate for the region is 633 mm, the average annual air temperature is + 6.7 °C (Figs 1, 2).

In some years significant fluctuations in moisture supply, both in the direction of excessive moisture and in the direction of precipitation deficit during the period of research under the general increase in temperature regime with regard to the average long-term indicators were observed. The years of 2015 and 2016 were the most optimal in terms of moisture supply and temperature regime.

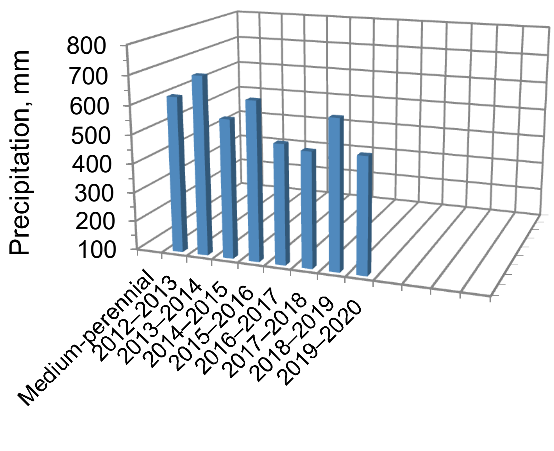

Figure 1. Precipitation (mm) for the period of research.

The soil of the experimental field was chernozem podzolized heavy loam low-humus on loess. The content of humus in the arable layer was 3.2−3.4%, the level of saturation of the bases in the range of 90−93%, the reaction of the soil solution was medium acid (pHKCl 5.7), hydrolytic acidity was 1.9−2.3 smmol kg^{-1} of soil, the content of mobile compounds of phosphorus and potassium (according to DSTU 4115-2002) was 125−150 mg kg^{-1}.

The initial material for hybridization was five species of winter hexaploid wheat, in particular, *Triticum spelta* L. (Zoria Ukrainy, Yevropa varieties), *Triticum sphaerococcum* Perciv. (Sharada variety), *Triticum aestivum* L. (Podolianka, Artemida, Freia, Artaplot varieties), *Triticum petropavlovskyi* Udacz., *Triticum compactum* Host.; samples of winter rye 193, 169 and 330 of Uman NUH selection and lines 308, 309 created at Nosivka selection-and-research station; samples of hexaploid triticale of own selection and varieties of Rozivska 6, Ladne, Khlibodar Kharkivsky, Beta, Alkid, Suvenir, Rarytet; samples of octaploid triticale UA0602463 and UA0601654 provided by the National center for plant genetic resources of Ukraine; wild forms of *Elimus arenarius* L.

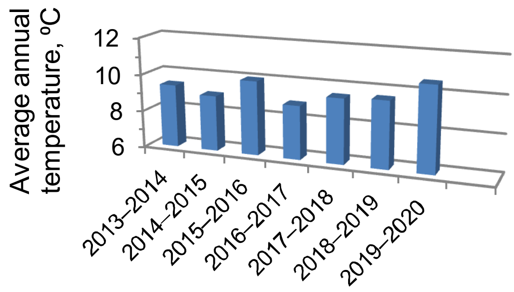

Figure 2. Average annual temperature (°C) for the period of research.

Hybridization was performed by emasculation (removal of anthers) of maternal flowers and their forced pollination by parental pollen form. After the seed maturing, the mother inflorescences were cut, the number of emasculated flowers and formed seed were counted, and the percentage of seed setting and the level of progamous incompatibility were determined. Pollen fertility was determined using dyes (methylene blue or iodine lugol solution). F_1 hybrids were sown in the field conditions and determined the field germination of seeds. $F_{2–5}$ hybrids were sown in four repetitions at the rate of 10 grains in a row of 1 m in length with a row spacing of 25 cm. The numbers were placed in blocks, where every 10^{th} number was the control variant. In the control varietal testing, the best samples were sown on the plots with the estimated area of 10 m^2 in four repetitions. A systematic method of plots placement was used in the research.

Hybrid progeny was analyzed by measuring morphological and economically valuable features, including plant height, lodging resistance, ear length, grain weight from the main ear and 1,000 grains weight, protein and gluten content in grain and its quality indicators, yield, etc. A systematic method of plots placement was used in the research. The numbers were arranged in blocks with a plant density of 400 thousand pcs ha^{-1} under four-time repetition. All accounting, observation and control variety testing of the created varieties were carried out in accordance with the 'Methodology of the State scientific-and-technical examination of plant varieties' (2012). State scientific-and-technical examination of the varieties of Navarra and Strateh was conducted during 2015–2018 at the State agricultural centers for examination of plant varieties in 17 regions of Ukraine. The reliability of research, the degree of features variation and the significance of differences between indicators of laboratory tests (protein and gluten content) were determined at the level of significance of $P \leq 0.01$, for field studies - $P \leq 0.05$ using statistical analysis program (SAS) v. 9.1.3. The variation coefficient

(V, %), standard deviation (S) and experimental error (Sx) were determined by the method of E. R. Ehrmantraut et al. (2000) and the use of MS Excel program.

RESULTS AND DISCUSSION

Hybridization of hexaploid wheat species with diploid rye. It was proved that combining disability due to the display of progamous incompatibility and death of hybrid embryos at different stages of their formation under intergeneric hybridization of wheat with rye was observed (Raina, 1984; Hills et al., 2007). This complicated the work of obtaining the primary forms of triticale.

It is known that soft wheat has genes that inhibit intergeneric compatibility, in particular, Kr_1 gene located in chromosome 5B, and Kr_2 gene located in chromosome 5A (Raina, 1984; Hua & Liu, 2012). Only 7% of the world's varieties of soft wheat cross well with rye (genotype $kr_1kr_1kr_2kr_2$, has setting of hybrid grains at the level of 50%), 14% - have a satisfactory level of compatibility (genotype $Kr_1Kr_1kr_2kr_2$, provides seed setting at the level of 10–30%, and genotype $kr_1kr_1Kr_2Kr_2$ - 30–50%), and 80% of varieties have low compatibility under crossing (genotype $Kr_1Kr_1Kr_2Kr_2$ – setting up to 10%. In addition, it is believed that there are other genes localized in D and R genomes that are responsible for intergeneric hybridization (Qi et al., 1999). The value of the cytoplasm is not eliminated (Hsam & Larter, 1974). Data on genetic control of species compatibility of *Triticum spelta* L., *Triticum compactum* Host. and *Triticum petropavlovskyi* Udacz. with rye in the scientific literature is not found. Since these species have identical genomic formula with soft wheat and chromosomal composition of individual subgenomes, it is possible to assume the presence of Kr-genes of different allelic state in their genome.

The success of crossing under remote hybridization depends on the level of progamic and postgamous incompatibility of the initial maternal and paternal forms. It can be determined in different ways, including: 1. By the ratio of the number of setting hybrid seed to the number of pollinated flowers. This gives an idea of the level of progamic incompatibility of the original forms, which varies significantly and depends not only on genotypes, but also on the technique of crossing, pollination time, pollen quality, weather conditions, etc. (Sechniak & Sulyma, 1984). 2. By the similarity of hybrid grains (the ratio of the number of similar seed to the total number of formed seed, expressed in a percentage), which characterizes the level of postgamous incompatibility of the original forms. This indicator varies less and depends on the parents' genotypes (Sechniak & Sulyma, 1984). 3. By the ratio of the number of similar hybrid grains to the total number of pollinated flowers, expressed in a percentage (Sechniak & Sulyma, 1984). 4. By the ratio of the number of hybrid plants that survived at the time of harvest to the number of wheat flowers pollinated with rye, expressed in a percentage (Manickavelu et al., 2009).

In the course of research, the analysis of the level of progamic and postgamous incompatibility of different types of hexaploid wheat under hybridization with diploid rye was carried out. The first and fourth methods to analyze the success of crossing were used. In 2015, samples of winter rye 193 and 169 were the pollinators. Spelt wheat of Zoria Ukrainy variety had a low level of seed setting under intergeneric hybridization, which indicates the presence of dominant alleles of incompatibility genes in the genome (Table 1).

Table 1. The level of seed setting under hybridization of different types of hexaploid wheat with rye, 2015–2017

Maternal form (wheat)			Paternal form (rye)			
Species	Variety	Probable combination of wheat genes	Level of seed setting, %	Survival of hybrid plants, %	Level of seed setting, %	Survival of hybrid plants, %
2015			Sample 193		Sample 169	
Triticum spelta L.	Zoria Ukrainy	$Kr_1Kr_1Kr_2Kr_2$	8.8	0.0	9.5	0.0
	Yevropa	$Kr_1Kr_1kr_2kr_2$	15.2	1.9	18.2	0.0
Triticum sphaerococcum Perciv.	Sharada	$Kr_1Kr_1kr_2kr_2$	18.7	0.0	21.2*	0.0
Triticum aestivum L.	Freia	$kr_1kr_1Kr_2Kr_2$	31.1*	10.7*	35.2*	9.5*
	Artaplot	$Kr_1Kr_1kr_2kr_2$	20.8	3.7	21.1*	0.0
2016			Line 308		Line 309	
Triticum spelta L.	Zoria Ukrainy	$Kr_1Kr_1Kr_2Kr_2$	6.5	0.0	4.2	0.0
	Yevropa	$Kr_1Kr_1kr_2kr_2$	12.8	4.7	13.8	4.9
Triticum sphaerococcum Perciv.	Sharada	$Kr_1Kr_1kr_2kr_2$	15.6	0.0	19.2	0.0
Triticum aestivum L.	Artemida	$kr_1kr_1Kr_2Kr_2$	32.5*	6.1	32.7*	5.8
	Podolianka	$Kr_1Kr_1kr_2kr_2$	25.8*	4.2	27.2*	4.1
Triticum petropavlovskyi Udacz.		$Kr_1Kr_1Kr_2Kr_2$	5.1	0.0	4.5	0.0
Triticum compactum Host.		$Kr_1Kr_1kr_2kr_2$	11.2	9.4	10.7	9.1
2017			Line 308		Line 330	
Triticum spelta L.	Zoria Ukrainy	Kr1Kr1Kr2Kr2	5.8	0.0	5.2	0.0
	Yevropa	Kr1Kr1kr2kr2	10.7	0.0	11.1	0.0
Triticum sphaerococcum Perciv.	Sharada	Kr1Kr1kr2kr2	18.2	0.0	20.2	0.0
Triticum petropavlovskyi Udacz.		Kr1Kr1Kr2Kr2	4.8	0.0	5.1	0.0
Triticum compactum Host.		Kr1Kr1kr2kr2	12.2	9.5*	12.8	9.7*

Note: * – the difference is relevant at the significance level of $P < 0.05$.

Spelt varieties of Yevropa, soft wheat of Artaplot and round grain wheat of Sharada had a higher level of compatibility with rye (setting in the range of 15.2–21.2%), which suggested the presence of kr_2 genes in the genome in a recessive homozygous state. The highest level of seed setting was recorded in soft winter wheat of Freia variety (over 30%), indicating its probable combination of $kr_1kr_1Kr_2Kr_2$ genes. The sample of winter rye 169 was more easily crossed with hexaploid wheat compared to sample 193. However, the higher combination ability was recorded when pollinating with sample 193.

In 2016, crossing of different types of hexaploid wheat was performed with winter rye lines 308 and 309, which were characterized by stunting (h = 72–74 cm), long ear (10–12 cm), and line 309 - by erectile arrangement of leaf blade. *Triticum petropavlovskyi* Udacz. species showed a low level of seed setting (4.5–5.1%) when crossing with diploid rye. Probably its genome is filled with dominant incompatibility genes. According to our data, spelt wheat variety of Zoria Ukrainy also had dominant incompatibility genes, as a small amount of seed (4.2–6.5%) was formed under hybridization with rye. However, under hybridization of spelt wheat variety of Yevropa with rye, a larger number of hybrid seed (12.8–13.8%) was obtained, which indicated the presence of the recessive kr_2 gene in the genome. This variety was created under hybridization of spelt with soft wheat (Polyanetska, 2012; Diordiieva et al., 2018a). Maybe, it inherited the recessive alleles of Kr_2/kr_2 gene from soft wheat. *Triticum compactum* Host. and *Triticum sphaerococcum* Perciv. species probably have $Kr_1Kr_1kr_2kr_2$ genotype, because seed setting under hybridization with rye was at the level of 10.7–21.2%. The highest rate of seed formation (32.5–32.7%) was recorded when crossing winter soft wheat of Artemida variety with rye. A possible combination of the variety genes was kr1kr1Kr2Kr2.

In 2017, winter rye line 308 and sample 330 which was created under hybridization of the maternal component of the hybrid by Dünger with self-pollinated line 149 were used as pollinators. The research results indicated the presence of dominant incompatibility genes in the genome of *Triticum petropavlovskyi* Udacz. species and in spelt wheat variety of Zoria Ukrainy, as seed setting for their crossing with rye was low. These results were confirmed by data from previous years. It was established the level of seed setting in the range of 10.7–20.2% for *Triticum compactum* Host., *Triticum sphaerococcum* Perciv. species and spelt wheat variety of Yevropa which indicated the presence of the recessive gene Kr_2/kr_2 in the genome.

Winter rye sample 330 was better crossed with hexaploid types of wheat than line 308 and provided a higher percentage of seed formation. Viable seed was obtained only in the combination of *Triticum compactum* Host. × sample 330 crossing, but hybrid plants could not be separated and propagated because hybrid generation did not set the seed even after hybridization with the fertile form of winter triticale.

Overcoming the sterility of wheat-and-rye F_1 hybrids. Wheat-and-rye F_1 hybrids are distinguished by sterility due to the lack of chromosome homology of the paternal species. Therefore, further work with them involves overcoming sterility. Currently, a number of ways to overcome the sterility of wheat-and-rye F_1 hybrids, in particular: 1) pollination of hybrid flowers with pollen of one of the paternal forms; 2) pollination of hybrid flowers with pollen of the third species with the same number of chromosomes; 3) method of meiotic polyploidization, which consists of pollination of hybrid flowers with pollen of hexaploid triticale; 4) providing optimal conditions for meiosis, gametogenesis, flowering, pollination and fertilization of hybrid plants (Grauda, 2010); 5) method of mitotic polyploidization, which consists of doubling the number of chromosomes in F_1 hybrids under the action of colchicine (Sechniak & Sulyma, 1984; Fu et al., 2010; Giacomin et al., 2015).

In our studies, seed obtained from plants of wheat-and-rye F_1 hybrids was sown on the plots. The method of meiotic polyploidization, which involves pollination of first-generation hybrids with pollen of hexaploid triticale, was chosen as the most optimal to overcome the pollen sterility. The chosen method allowed expanding the genetic diversity of hexaploid triticale. During such crosses, gradual elimination in hybrids of D genome of soft wheat and formation of constant genotypes with the genomic formula AABBRR were occurred, that is secondary hexaploid triticale was formed (Han et al., 2003). The recombination potential of triticale was wide because of the influence under its formation of soft and durum wheat species with genomes of different quality (Kwiatek et al., 2015; Diordiieva et al., 2018a; Kwiatek & Nawracała, 2018).

In order to overcome the sterility of plants in the remote F_1 hybrids, they were pollinated with pollen of different samples of hexaploid triticale. Some wheat-and-rye hybrids of the first generation were easily crossed with hexaploid forms (Freia × sample 169, *Triticum compactum* Host. × line 308), some of them did not cross at all, did not set seed, and, as a result, they eliminated (Yevropa × line 308, *Triticum compactum* Host. × line 309, Yevropa × line 309) (Table 2). Scientists (Sechniak & Sulyma, 1984) indicate a higher percentage of seed setting under spontaneous pollination of sterile remote hybrids. However, according to our research results, better setting was provided by controlled forced pollination. Ears that were not placed under the isolator for artificial pollination (50% of spikes of each plant) in the vast majority of cases did not form seed. As an exception, it was recorded only one case of seed setting under spontaneous plants pollination ♀ Yevropa variety × ♂ winter rye line 308 when two seeds were formed, and they turned out to be non-viable.

Table 2. Results of pollination of the obtained wheat-and-rye F_1 hybrids with pollen of hexaploid triticale, 2016–2018

Combination of crossing ♀ F_1 (wheat × winter rye) × ♂ 6x triticale	Number of, pcs.				Level of seed setting, %	Germination capacity, %
	Pollinated ears	Pollinated flowers	Formed seed	Obtained plants		
(Yevropa variety × sample 193) × Alkid variety	3	62	8	1	12.9	12.5
(Yevropa variety × line 308) × Suvenir variety	1	23	0	0	0.0	0.0
(Yevropa variety × line 309) × Suvenir variety	1	23	2	0	0.0	0.0
(Freia variety × sample 169) × Alkid variety	3	65	10	2	15.4	20.0
(Freia variety × sample 193) × pollen mixture of 6x triticale**	2	45	8	2	17.8*	25.0*
(Artaplot variety × sample 193) × Suvenir variety	1	27	0	0	0.0	0.0
(Artemida variety × line 308) × pollen mixture of 6x triticale	1	28	4	1	14.3	25.0
(Artemida variety × line 309) × Alkid variety	1	28	3	0	0.0	0.0
(Podolianka variety × line 308) × pollen mixture of 6x triticale	1	27	6	1	22.2*	16.7
(Podolianka variety × line 309) × pollen mixture of 6x triticale	1	26	6	2	23.1*	33.3*
(*Triticum compactum* Host. × line 308) × Alkid variety	2	65	10	2	15.4	20.0
(*Triticum compactum* Host. × line 309) × pollen mixture of 6x triticale	1	23	0	0	0.0	0.0
(*Triticum compactum* Host. × sample 330) × pollen mixture of 6x triticale	1	24	5	1	20.8*	20.0

Note: * – the difference is relevant at the significance level of $P \leq 0.05$;
** the pollen donors were varieties of winter triticale Alkid and Suvenir at the ratio of 50:50.

Increase in the level of seed setting under pollination of wheat-and-rye F_1 hybrids was observed. In hybrids, the influence of incompatibility genes disappears, which probably leads to the increase in the level of seed setting. The largest number of grains

(10 pieces) was formed by F_1 hybrids obtained in the combination of crossing (Freia × sample 169) × Alkid and (*Triticum compactum* Host. × line 308) × Alkid. Hybrid seed had low germination capacity (in most cases, one or two seeds were viable).

Seed setting and its field germination increased under saturating crosses of wheat-and-rye F_2 hybrids with pollen of hexaploid triticale.

In addition, in three combinations of crossing, seed formation as a result of free pollination by an unknown parental form was recorded. At the same time, the level of seed setting under free pollination was 5–10% higher than under controlled crossing. In plants obtained from the pollination of wheat-and-rye F_2 hybrids with pollen of hexaploid triticale, restoration of pollen fertility was usually observed. In the future, individual selection and analysis of elite plants among the obtained samples, which will allow choosing a valuable original material and sources of economically valuable features for selection improvement of hexaploid triticale, can be carried out.

Hybridization of octa- and hexaploid triticale with spelt wheat and *Elimus arenarius* L. The compatibility of triticale with wheat, in particular spelt, is determined by the genotypes of the original forms included in the crossing scheme, and depends on meteorological conditions (Li et al., 2015). A broad formative process in hybrids of hexaploid triticale with spelt wheat was observed; in the result of which recombinant forms that can be used in the selection work by genes donors of economically valuable features were formed (Diordiieva et al., 2018b).

It was found that seed setting under triticale crossing with spelt wheat was low, regardless of the octaploid (2.8–3.2%) or hexaploid (1.2–8.0%) level of crop ploidy (Table 3).

Table 3. Level of seed setting under crossing octa- and hexaploid forms of triticale with spelt wheat and *Elimus arenarius* L., 2014

Maternal form		Paternal form	Number of, pcs. Castrated flowers	Formed grains	Level of seed setting, %
Octaploid forms	UA0602463*,	Zoria Ukrainy variety	687	22**	3.2
	UA0601654*	Zoria Ukrainy variety	632	18**	2.8
	UA0602463*,	*Elimus arenarius*	250	3	1.2
Hexaploid forms	Rozivska variety 6	Zoria Ukrainy variety	350	16	4.6
	Ladne variety	Zoria Ukrainy variety	324	17**	5.2**
	Rarytet variety	Zoria Ukrainy variety	287	14	4.9
	Rarytet variety	*Elimus arenarius*	285	4	1.4
	Suvenir variety	Zoria Ukrainy variety	315	18	5.7**
	Khibodar kharkivsky variety	Zoria Ukrainy variety	452	20**	4.4
	Alkid variety	Zoria Ukrainy variety	560	22**	3.9
	Alkid variety	*Elimus arenarius*	290	6	2.1
	Beta variety	Zoria Ukrainy variety	187	15	8.0**

Note: * – sample number according to the catalogue of the National Center for Plant Genetic Resources of Ukraine; ** – the difference is relevant at the significance level of $P \leq 0.05$.

Number of researches showed that the lack of homologous conjugation between chromosomes of the genomes *R* of triticale and *D* of wheat leads to abnormalities in the embryonic development of hybrids (Badaev et al., 1985; Khanna, 1990; Kalinka & Achrem, 2018).

We can expect the same deviations under hybridization of triticale with spelt wheat because spelt wheat has a genomic composition similar to soft wheat. As a result, deformed and thin grains with different shapes of hybrid caryopsis were formed.

F_1 hybrids from crossing hexaploid triticale with spelt by the morphological structure of the ear and the general habit of plants are of the same type. Characteristic features of F_1 hybrids were the presence of a long fluffy bald ear, coarse glume and complicated grain threshing, which is probably inherited from spelt wheat. F_1 hybrids from crossing octaploid triticale with spelt by the morphology of plants and ears approached wheat, because their genotype was quantitatively dominated by wheat genomes in a ratio of 3:1, which was qualitatively supplemented by the spelt genetic material. Their ear is bald or semi-bald, of medium length (11–13 cm) and density.

Awnless of hexaploid wheat species is controlled by *B1, B2, Hd* genes which are localized in *5A, 6B* and *4A* chromosomes. Baldness in first-generation hybrids while crossing barbate hexaploid triticale with bald forms of soft wheat is dominated. In subsequent generations, the expression of the baldness feature is significantly reduced to its complete absence. The reasons for such changes have not been clearly determined (Johnson et al., 2008). Spelt wheat contains a recessive allele *q* in a homozygous state, which controls the formation of the speltoid type of ear, and the dominant *TgTg* alleles, which control the presence of coarse glume. The presence of at least one of these alleles leads to ear elongation and complication of threshing in hybrid offspring (De Faris et al., 2006). Hybrids of triticale × spelt wheat are heterozygous by these alleles in the first generation, which explains the homogeneity of the morphological structure of the ear and the phenotypic manifestation of spelt wheat features.

Hybridization of triticale with *Elimus arenarius* L. provided a lower level of seed setting, that was 1.2%, for crossing combinations with octaploid forms, and - 1.4–2.1% with hexaploid ones. Hybrids obtained under the participation of *Elimus arenarius* L. were characterized by a long (20–22 cm) ear with a significant number of spikelets and flowers. The phenotype of the plants was similar to triticale. The presence of a waxy coating and dove-colored plants, which, apparently, was inherited from *Elimus arenarius* L. was their distinct difference from the original varieties of triticale. Triticale hybrids obtained under the participation of spelt wheat and *Elimus arenarius* L. were sterile. Only a few cases of formation of fertile pollen grains were recorded. Spontaneous pollination of the first generation hybrids gave low indicators of ear grain content of F_1 plants, which was 0.0–2.5% for octaploid triticale and 0.0–3.0% for hexaploid triticale (Table 4).

It was observed a higher level of seed setting under isolation and artificial pollination of ears of the F_1 hybrid plants obtained under the participation of spelt wheat with hexaploid triticale pollen, which was 5.2–8.7% for crossing combinations with octaploid triticale, and 3.5–9.0% with hexaploid one. Seed did not set under artificial pollination of F_1 hybrids (octaploid triticale × *Elimus arenarius* L.) with pollen of the maternal seed form. Six seeds under hybrids re-crossing (hexaploid triticale × *Elimus arenarius* L.) with hexaploid triticale were obtained, but only one of them was germinable.

Table 4. Level of seed setting of remote hybrids under spontaneous and artificial pollination, 2015

Crossing combination	Spontaneous pollination			Artificial pollination with pollen of hexaploid triticale		
	Number of formed seed, pcs.	Level of seed Setting, %	Germination Capacity, %	Number of formed seed, pcs.	Level of seed setting, %	Germination Capacity, %
♀ UA0602463 × ♂ Zoria Ukrainy variety	4	2.5	0.0	12	5.2	16.7*
♀ UA0601654 × ♂ Zoria Ukrainy variety	0	0.0	0.0	14*	8.7*	14.3
♀ UA0602463 × ♂*Elimus arenarius* L.	0	0.0	0.0	0	0.0	0.0
♀ Rozivska variety 6 × ♂ Zoria Ukrainy variety	7	2.7*	14.3	8	3.5	0.0
♀ Ladne variety × ♂ Zoria Ukrainy variety	0	0.0	0.0	9	6.0	11.0
♀ Rarytet variety × ♂ Zoria Ukrainy variety	6	2.1	16.7	10	7.1	10.0
♀ Rarytet variety × ♂ *Elimus arenarius* L.	0	0.0	0.0	2	1.1	0.0
♀ Suvenir variety × ♂ Zoria Ukrainy variety	8	3.0*	25.0*	12	7.5	16.7
♀ Khlibodar kharkivsky variety × ♂ Zoria Ukrainy variety	6	2.1	0.0	14*	8.8*	7.1
♀ Alkid variety × ♂ Zoria Ukrainy variety	9	2.5	11.0	14*	9.0*	14.2
♀ Alkid variety × ♂ *Elimus arenarius* L.	0	0.0	0.0	4	1.5	25.0*
♀ Beta variety × ♂ Zoria Ukrainy variety	8	2.7*	25.0*	10	8.5*	10.0

Note: * – the difference is relevant at the significance level of $P \leq 0.05$.

Seeds obtained by both methods of pollination (artificial and spontaneous) were thin and deformed. Most grains were not viable, the germination capacity ranged from 0.0 to 25.0% Octa- and hexaploid triticale (genomic formula, ABDR and ABR, respectively), although they have genomes related to spelt wheat (genomic formula ABD), but they differ in the presence of rye genome R. This leads to a lack of homologous conjugation between the chromosomes of the parental forms causing abnormalities in embryonic development in hybrids. A wider range of abnormalities in embryonic development in hybrids under hybridization of triticale with *Elimus arenarius* L. can be observed, because these species do not have related genomes at all. This explains the formation of deformed and slender seed with different forms of hybrid caryopses and low viability.

Further work with the obtained hybrids consisted in backcrossing with their pollen of certain varieties of triticale and resowing of the obtained seed. Plants of the formed populations differed in height, morphological features of an ear, pollen fertility, etc. They were characterized by a high level of pollen sterility, which led to a small number of productive plants.

Analysis of triticale samples by indicators of grain productivity and quality. New samples of winter triticale were synthesized under hybridization of octa- and hexaploid triticale with spelt wheat and *Elimus arenarius* L., different types of wheat with winter rye. A number of new materials due to the intensive form-building process, which were analyzed by morphobiological properties and economically valuable features, were obtained. As a result of research, it was identified samples that exceeded the standard in terms of yield and elements of ear productivity.

Breeding materials obtained with the participation of spelt wheat were characterized by a significant range of variability in plant height (V = 25%) (Table 5). According to this indicator, all samples were divided into short-stemmed (h = 60–80 cm), low-stemmed (h = 81–100 cm) and medium-stemmed (h = more than 100 cm). All samples obtained with the participation of *Elimus arenarius* L. were classified as medium-stemmed (h = 110–118 cm).

Table 5. Indicators of economically valuable features of winter triticale samples created under remote hybridization, 2018–2020

Breedind material	Plant height, cm	Lodging		Ear length, cm	Grain mass from the ear, gr	Yield, t ha^{-1}	1,000 seeds mass, gr	Content of, %	
		%	Resistance points					Gluten	Protein
Group standard*	110	4	8	12.2	2.03	6.48	48.5	24.2	11.8
Samples obtained with participation of spelt wheat									
28	112	12	6	14.2	2.08	5.87	49.1	26.6**	13.0**
35	115	4	8	13.2	2.35**	6.81**	48.5	23.8	12.4**
61	95	3	8	12.0	2.10	6.92	49.4	26.0**	12.4**
68	87	3	8	12.8**	2.22**	6.95**	50.5**	27.8**	12.8**
85	118**	8	7	12.5	2.01	5.77	45.1	26.8**	12.5**
92	110	6	7	13.0**	1.87	5.92	48.7	25.4**	12.2**
112	108	2	8	14.1	2.12**	6.95**	46.8	23.8	12.2**
254	92	15	5	13.2	1.75	5.56	47.2	22.4	11.5
455	108**	3	8	12.0	1.98	6.15	48.0	30.2**	14.2**
481	87	3	8	12.8**	2.20**	6.85**	48.2	20.8	11.1
484	85	5	7	12.5	2.18	6.74	50.2**	21.9	11.5
491	110	5	7	13.4**	2.25**	6.53	48.2	26.4**	12.5**
x	102	–	–	12.9	2.09	6.42	48.3	25.2	12.4
S	12	–	–	0.7	0.17	0.53	1.5	2.7	0.8
$LSD_{0.95}$	*4*	–	–	*0.4*	*0.09*	*0.27*	*1.6*	*0.2*	*0.1*
V, %	*25*	–	–	*8*	*12*	*18*	*13*	*8*	*6*
Sx, %	*4*	–	–	*3.7*	*4.2*	*4.3*	*3.5*	*0.8*	*0.9*
Samples obtained with participation of *Elimus arenarius* L.									
5	118**	10	6	20.8**	1.58	5.35	42.2	22.1	11.1
8	115**	8	7	22.4**	1.75	5.58	43.1	23.6	11.6
10	110	12	6	19.5**	1.62	5.41	40.8	22.5	11.2
13	114	13	6	20.2**	1.47	5.18	41.2	22.8	11.4
x	114	–	–	20.7	1.61	5.38	41.8	22.8	11.3
S	3	–	–	1.2	0.12	0.17	1.0	0.6	0.2
$LSD_{0.95}$	*4*	–	–	*0.7*	*0.06*	*0.20*	*1.6*	*0.2*	*0.1*
V, %	*9*	–	–	*11*	*15*	*16*	*7*	*10*	*8*
Sx, %	*4*	–	–	*3.5*	*4.0*	*4.1*	*3.7*	*0.8*	*0.9*

Note. *Group standard – winter triticale varieties of Alkid, Rarytet, Suvenir; ** – the difference is relevant, higher than group standard at the significance level of $P \leq 0.05$.

Species *Triticum spelta* L. and *Elimus arenarius* L. were characterized by a long ear (*Triticum spelta* – up to 20 cm, *Elimus arenarius* L. - up to 30 cm) (Gabel, 1984; Gulyas et al., 2012). Hybridization of three-species triticale with these species led to ear elongation in the offspring. In such a case, crossing with spelt caused a slight (0.4–1.9 cm) ear elongation, and hybridization with *Elimus arenarius* L. contributed to the production of offspring with an ear length in the range of 19.5–22.4 cm, which significantly exceeded the group standard and samples obtained with the participation of spelt wheat.

However, the materials obtained under hybridization of triticale with *Elimus arenarius* L. were characterized by a decrease in grain mass from the ear and a decrease in yield compared to the group standard. There were identified seven samples (35, 61, 68, 481, 484 and 491) among the ones obtained with the participation of spelt wheat, which significantly exceeded the group standard by yielding capacity.

It was found that hybridization of three-species triticale with spelt wheat had a positive effect on the indicators of offspring grain quality, in particular, on the protein and gluten content.

Nine of the 12 samples that had spelt in the generation, significantly exceeded the group standard by protein content in the grain, seven of which were characterized by a significant increase in gluten content and two -by the essential increase in 1,000 seed mass. Increase in protein and gluten content in grain indicates a positive effect of crossing triticale with spelt wheat, which is characterized by a high level of these indicators manifestation (protein - up to 25%, gluten - up to 50%) and a high coefficient of inheritance of these criteria in hybrids. Hybridization of these two species was carried out in order to create new triticale with high grain quality.

The great weight of 1,000 grains in sample 484 is obviously inherited from the maternal form of the variety of three-species triticale Alkid, which is one of the most productive in Ukraine and is characterized by a weight of 1,000 grains at the level of 50.5–51.0 g. Hybridization of three-species triticale with *Elimus arenarius* L. led to the great decrease in all grain quality indicators in the offspring, which was expected because *Elimus arenarius* L. is a wild species that has no production value. It was involved in the hybridization system as a donor of high indicators of ear productivity.

The results of winter triticale breeding. Hexaploid winter triticale varieties of Navarra and Strateh in the result of hybridization of three-species triticale and spelt wheat and multiple individual selections were created, and they were included in the State register of plant varieties suitable for distribution in Ukraine since 2018.

Varieties by the method of remote hybridization of three-species forms of triticale and spelt wheat, followed by individual selections in $F_{2–4}$ and repeated improving selections in terms of productivity and grain quality in $F_{5–6}$ were created. Navarra variety (sample 491) was created by hybridization of three-species triticale of Rozivska 6 variety with a sample of spelt wheat from the submontane regions of the Carpathians, followed by crossing of the first generation hybrids with spring triticale variety Khlibodar Kharkivsky. Strateh variety (sample 455 awnless) was created by crossing winter triticale varieties Rozivska 6 and Alkid and pollination of the first generation hybrid with pollen of spelt wheat sample from the submontane regions of the Carpathians. During their creation, an important task was set - to increase protein and gluten content in grain due to the introgression of genetic material of spelt wheat into the hexaploid triticale genotype. Their genotype combined the genetic material of the varieties of three-species triticale, which was created in selection institutions located in remote ecological-and-geographical areas of Ukraine. The best samples analyzed in the selection nursery by displaying of economically valuable features with the help of multiple individual selections were chosen. Five numbers after a severe rejection of families in terms of productivity, grain quality and lodging resistance were chosen. After materials approbation, high-yielding samples 491 and 455 (awnless), which were analyzed in a competitive variety testing, were separated. These samples were selected as constant genotypes characterized by high yields (sample 491) and high protein and gluten content (sample 455). In addition, sample 455 was characterized by awnless, which was quite rare in winter triticale. Sample 491, except for spelt wheat, contains the genetic material of triticale of different developmental types (winter and spring) in its genome.

In the course of research it was established that the average yield of 491 sample for the period of competitive variety testing (2013–2015) in the conditions of Uman NUH was 5.97 t ha^{-1}, which significantly exceeded the group standard (Table 6). The average yield of sample 455 (awnless) was 4.96 t ha^{-1}, which was not significantly inferior to the group standard. Sample 491 was characterized by a high level of displaying of economically valuable features; in particular, it was significantly inferior to the group standard by the plants height (102 cm) and significantly exceeded it in terms of lodging resistance. It did not differ significantly from the control variant indicators in terms of grain quality (gluten content - 21.7%, grain unit - 690 g L^{-1}, 1,000 seeds mass - 47.8 g).

Table 6. Productivity indicators of winter triticale samples 491 and 455 (awnless) under competitive variety testing in the conditions of Uman NUH, 2013–2015

Indicators	Group standard*	Genotype (factor A)		*LsD*$_{0.95}$		
		Sample 491	Sample 455	A	B	AB
Year (factor B) 2013						
Yield, t ha^{-1}	5.02	5.75**	4.78	0.12***	0.09	0.19
				30.0	22.5	47.5
Plant height, cm	108	100	103	4.12	1.25	3.25
				47.8	14.5	37.7
Lodging, %	19.8	5.4	32.1	–		
resistance points	5	7	5			
Gluten content, %	21.5	21.7	29.8**	0.65	0.15	0.68
				43.9	10.2	45.9
Grain unit, g L^{-1}	685	690	685	21.2	12.2	30.8
				33.0	18.9	48.0
1,000 seed mass, g	47.1	47.1	46.2	2.12	0.74	2.35
				40.7	14.2	45.1
2014						
Yield, t ha^{-1}	5.35	6.12**	5.14	0.14	0.12	0.21
				29.8	25.5	44.7
Plant height, cm	112	104	109	4.25	1.28	3.58
				46.7	14.1	39.3
Lodging, %	29.1	10.2	48.5	–		
resistance points	5	6	3			
Gluten content, %	20.1	20.5	28.1**	0.58	0.21	0.75
				37.7	13.6	48.7
Grain unit, g L^{-1}	680	680	680	20.1	12.1	30.2
				32.2	19.4	48.4
1,000 seed mass, g	48.5	48.6	45.4	2.15	1.28	2.40
				36.9	21.9	41.2

Table 6 (continued)

2015						
Yield, t ha^{-1}	5.15	6.04**	4.96	0.12	0.10	0.20
				28.6	23.8	47.6
Plant height, cm	110	102	108	4.18	1.25	3.47
				47.0	14.0	39.0
Lodging, %	26.4	8.9	43.9	–		
resistance points	5	7	3			
Gluten content, %	22.8	22.8	31.4**	0.61	0.22	0.71
				37.2	13.4	49.4
Grain unit, g L^{-1}	690	700	690	20.5	11.8	30.5
				32.6	18.8	48.6
1,000 seed mass, g	48.0	47.8	47.5	2.21	1.21	2.75
				35.8	19.6	44.6

Note: *Group standard – varieties of winter triticale Alkid, Rarytet, Suvenir; ** – the difference is relevant, higher than standard at the significance level of $P < 0.05$; *** above the line – LsD_{05} by factors, under the line – % influence of the factor on the feature manifestation.

Dispersion analysis showed relevant differences between genotypes, environments and their interaction by all features, as well as the difference in the influence of these factors on the formation of the level of features manifestation. Thus, the main factor in yield dispersion, gluten content, grain unit and mass of 1,000 seed was the interaction of genotype-environment. Genotype determined dispersions of plant height throughout all years of research.

According to the results of a three-year competitive variety testing, sample 491 in 2015 was submitted to the State scientific-and-technical examination under the name of Navarra variety. Sample 455 (awnless) successfully combined high yield and high indicators of grain quality (gluten content - 29.8%, protein - 13.0%, grain unit - 685 g L^{-1}). 1,000 seeds mass was 48.2 g. According to the results of a three-year competitive variety testing, sample 455 (awnless) in 2015 was submitted to the State scientific-and-technical examination under the name of Strateh variety.

The state scientific-and-technical examination of the created varieties passed during 2015–2018 in 17 regional State centers of plant varieties examination. During this period, the average yield of Navarra variety in the Polissia zone was 5.46 t ha^{-1}, which exceeded the zone average indicators by 1.0 t ha^{-1} (Table 7).

Navarra variety was characterized by high grain quality, in particular, by protein content - 13.1%, 1,000 seeds mass - 48.6 g and was characterized by high resistance (8.3–9.0 points) to adverse environmental factors (fall, drought, fungal diseases). A slight lodging (resistance point - 6.5) should be noted among the negative characteristics of the variety.

The variety yield in the Forest-Steppe zone was lower and was to 5.29 t ha$^{-1.}$ However, it should be mentioned its higher resistance to adverse biotic and abiotic environmental factors, which was not inferior to the group standard in this area (8.5–9.0 points). It should be noted that a decrease in plant height to 94 cm in the Forest-Steppe zone in Navarra variety compared to the same indicator in the Polissia zone (114 cm) was recorded. This had a positive effect on its resistance to lodging (7.5 points).

Table 7. Productivity indicators of winter triticale variety of Navarra according to the results of the State scientific-and-technical examination, 2015–2018

Indicator		Forest-Steppe			Polissia		
		Average by zone	Navarra	Strateh	Average by zone	Navarra	Strateh
Yield, t ha^{-1}		5.59	5.26	5.13	4.46	5.46*	5.03
Resistance (points) to	Fall	8.9	9.1	9.1	9.1	8.8	8.8
	Drought	8.5	8.5	8.5	8.5	8.8	8.5
	Lodging	8.1	7.5	8.6	8.6	6.5	7.0
	root rot	9.0	9.0	9.1	9.1	9.0	9.0
	Fusariosis	9.0	9.0	9.1	9.1	8.5	8.5
	powdery mildew	9.0	9.0	9.1	9.1	9.0	9.0
	brown rust	9.0	9.0	9.1	9.1	8.3	8.8
Plants height, cm		104	94	109	113	114	140*
Unseeded ear, %		12.7	20.0	19.0	16.6	12.3	14.7
1,000 seeds mass, g		47.8	48.3	49.2	46.6	48.6	48.9*
Protein content, %		–	13.0	14.3*	–	13.1	14.0*

Note: * – the difference is relevantly higher than the average by the indicators area at the significance level of $P < 0.05$.

The yield of Strateh variety in the Polissia zone averaged 5.03 t ha^{-1} which exceeded average indicators by zone by 0.57 t ha^{-1}. The variety was characterized by high grain quality, by protein content - 14.0%. It had a complex high resistance (8.5–9.0 points) to adverse environmental factors and it was characterized by low plants lodging (7.0 points), which was obviously due to the average height of the plant stand of 140 cm. 1,000 seeds mass - 48.9 g. The yield of Strateh variety (5.13 t ha^{-1}) in the Forest-Steppe zone was lower in comparison with the average indicators of the zone (5.59 t ha$^{-1)}$ However, the variety was characterized by a high content of protein in grain (14.3%) and an average plant height of 109 cm, which had a positive effect on lodging resistance (8.6 points).

According to the results of the State scientific-and-technical examination, Navarra and Strateh varieties were included in the State register of plant varieties suitable for distribution in Ukraine in 2018 and recommended for cultivation in the Polissia zone.

CONCLUSIONS

1. It is proved that the species *Triticum spelta* L. and *Triticum petropavlovskyi* Udacz. had dominant genes of incompatibility with rye, their hybridization and seed formation process was difficult, and hybrid seed did not germinate in the field conditions. Crossing *Triticum compactum* Host. and *Triticum sphaerococcum* Perciv. species with rye allowed obtaining a higher level of seed setting, but the grain obtained with the participation of *Triticum sphaerococcum* Perciv. species was not viable. Pollination of remote wheat-and-rye hybrids with hexaploid triticale pollen led to partial stabilization of the chromosome set and restoration of pollen fertility.

2. Compatibility of triticale with spelt wheat and *Elimus arenarius* L. was low regardless of the level of triticale ploidy. The level of seed formation in F_1 hybrids under artificial pollination was higher than under spontaneous pollination. Seed germination

obtained from pollination of F_1 hybrids by fertile forms of triticale, regardless of the method of pollination and pollinator - was low.

3. It was found that hybridization of three-species triticale with spelt wheat had a positive effect on grain quality indicators in the offspring, in particular, on protein and gluten content. Crossing triticale with *Elimus arenarius* L. led to ear elongation, but caused a significant reduction in all indicators of grain quality in the offspring.

4. As a result of remote hybridization of three-species triticale and spelt wheat, winter triticale varieties of Navarra and Strateh were created, which were included in the State register of plant varieties suitable for distribution in Ukraine since 2018.

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