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RESEARCH ON THE VARIABILITY IN TRITICALE (*xTRITICOSECALE* WITTM.) CROSSES AS A SOURCE OF GENETIC DIVERSITY

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Abstract: In order to determine the possibility of occurrence of higher genetic diversity in hybrid combinations, ten triticale crosses in first hybrid generation and eleven triticale varieties were studied. Complete variation analysis of the studied parameters was carried out. Crosses 24/14 (Akord x Bumerang), 28/14 (Akord x Doni 52), 35/14 (Respekt x Irnik) and 36/14 (Respekt x Dobrudzhanets) demonstrated high variation of most of the parameters. Very low variation for a large number of indices was found in crosses 12/14 (Atila x Akord), 13/14 (Atila x Respekt), 26/14 (Akord x Dobrudzhanets) and 34/14 (Respekt x Bumerang). In combination with the mean values of the respective parameters, crosses 28/14 (Akord x Doni 52) and 36/14 (Respekt x Dobrudzhanets) were with highest probability for greater genetic diversity, which makes them valuable breeding material in the improvement work on triticale.

INTRODUCTION

The biological diversity in the agroecosystem is characterized with extremely low values due to the predominance of a particular single species – the cultural plant (Swift et al., 1996). Although such a biological system is not lacking biodiversity in the absolute sense, the rest of the species are dramatically less in number, which implies shift in the ecological balance (Altieri, 1999). On the other hand, the cultural plant, represented by a specific cultivar or hybrid, should not be regarded as a narrow-sense completely uniform biological system, but as a system with its own diversity (Hajjar et al., 2008).

Developing of cultivars in the self pollinated cultural plants is often related to the pure line theory of Johansson (Chahal and Gossal, 2000). In reality, the developing (breeding) of a pure line in the narrow sense is practically impossible because uniformity according to an enormous number of genes has to be achieved (Agrawal, 1998; Frey, 1983; Genchev et al., 1961; Acquaah, 2007). In practice, the cultivars consist of mixed populations where the plants are uniform only and exclusively according to the desired breeding characteristics (Genchev et al., 1961; Chahal and Gossal, 2000). In this respect, it should be pointed out that each cultivar or hybrid, regardless of its complete theoretical uniformity, is characterized with specific genetic variability. This variability allows the plant sets to adapt to the changeable environment, a specific gene expression being predominant accordingly. This gene expression is manifested as morphological diversity in the individual components of the population, which comprises the variety of the hybrid (Raser and O'Shea, 2005). Therefore, the morphological diversity has to be in high and significant correlation with the genetic diversity. A basic parameter for evaluation of the morphological diversity (or of the genetic one) is the evaluation of the variation of certain morphological indices (Sabaghnia et al., 2014; Idi Garba et al., 2015; Brezinova et al., 2009).

Triticale, as a product of distant hybridization and a self-pollinating crop, has its peculiarities. On the one hand, full uniformity of lines is extremely difficult to achieve with the cultivars because, due to their amphiploid nature, complex genetic inter-genomic interactions are observed, which are related to great deviations in the breeding indices (Baychev, 1990; Zumelzu et al., 1992; Ordonez et al., 1997). This implies long-term selection and high breeding pressure in order to achieve the desired uniformity of the breeding characteristics (Randhawa et al., 2015; Sechnyak and Sulima, 1984). Nevertheless, in the developed lines (in broad sense) and cultivars, full uniformity is not observed and triticale is often characterized with higher variability of the yield's structural elements in comparison to the other cereals (Sechnyak and Sulima, 1984; Baychev and Mihova, 2014).

On the other hand, it is possible to observe strong deviations in the triticale hybrid combinations even in the early generations of the hybrid offpsring due to the complex interactions between the sub genomes of the two parental forms (Lelley, 2006). Therefore, in spite of the theoretical expectations for high similarity between the individual plants, sharp deviations in the structural elements of yield may occur (Baychev, 1996). Regardless of that, the early generations of the crosses possess unique genome with specific response to the environment (Oettler et al., 2004). Since each deviation from the meteorological conditions, averaged for a certain region, is considered stressful for the crop, the assessment of the variation is related to the range of the reaction to stress, or to the ecological plasticity and stability of a given genotype. Thus the variation of the structural elements of spike, both in the cultivars and the hybrid combinations between them, is indicative for their genetic variability. The evaluation of only the hybrid combinations, without taking into account the data on the parental components and the similar varieties, cannot give a clear idea about the genetic diversity in the crop. Therefore, it is necessary to analyze the first hybrid generation as well as the parental forms.

The aim of this paper, based on the evaluation of the variation in the cultivars and in the first-generation hybrid combinations of triticale, was to assess their genetic variability, related to the breeding program of this crop.

MATERIALS AND METHODS

Plant material

Eleven Bulgarian triticale cultivars were used in the present study (Table 1).

Table 1. Triticale cultivars used in the investigation

No	Name	Origin	Year of registration
1	Kolorit	BGL “S” – BGC / 568-343	2005
2	Atila	AD 8x(Ep 1034/79 x Harkovska 60) / F_1[F_1(Yuzhnaya zrya/ Harkovska 60) / 804-503]	2007
3	Akord	MT-3 / F_2 populations	2007
4	Respekt	1262-12-2-10 / Veleten	2008
5	Bumerang	LP 3090.91 / 2853-1044	2009
6	Irnik	5252 - 131 / 2853-1044	2011
7	Dobrudzhanets	Chrono / 2853-1044	2012
8	Lovchanets	F_1 (Tornado / 3493-699) / Zaryad	2013
9	Doni 52	5279-131 / 3370-190	2014
10	Blagovest	32/99 / Zaryad	2015
11	Borislav	46/95-96 / 129/98	2016

Ten crosses were also included, in which one of the parents was highly cold resistant (Akord and Respekt) (Baychev, 2013), while the other was highly drought resistant (Atila, Bumerang, Doni 52, Irnik and Dobrudzhanets, preliminary data). These crosses are presented in Table 2.

Experimental design

The experiment was carried out during the year 2014/2015. The first hybrid generation of the used crosses was planted in plots of 2 m rows, with 30 cm interspacing and 10 cm distance between the plants in the rows. The number of rows of each hybrid combination depend on the number of seeds obtained from the cross. Planting was manual, within the sowing dates standard for triticale (10^{th} – 15^{th} October), using 20 grains per row. During the growth season, the indices date to heading (DH) and plant height (PH, measured from the base of the plant to the top of the spike, without the awns) were evaluated in the entire plant set and in the crosses. Harvesting was carried out at full maturity. The main spike from each plant was analyzed according to the characteristic spike length (SL, cm), number of spikelets per spike (NSS), number of grains per spike (NGS), grain weight per spike (GWS, g), 1000 kernel weight, TKW, g), spike density (number of spikelets per 1 cm of spike length) (SD) – NSS to SL ratio, fertility of spikelet (number of grains per spikelet) (F) – NGS to NSS ratio.

The cultivars used as parental forms and the other triticale cultivars were planted, grown and analyzed in identical manner. The analysis was carried out according to the same characteristics as used in the hybrid progenies.

Table 2. Origin of the used crosses

No	Breeding No	Origin
1	12/14	Atila x Akord (At x Ak)*
2	13/14	Atila x Respekt (At x R)
3	24/14	Akord x Bumerang (Ak x B)
4	25/14	Akord x Irnik (Ak x I)
5	26/14	Akord x Dobrudzhanets (Ak x Dr)
6	28/14	Akord x Doni 52 (Ak x Dn)
7	34/14	Respekt x Bumerang (R x B)
8	35/14	Respekt x Irnik (R x I)
9	36/14	Respekt x Dobrudzhanets (R x Dr)
10	38/14	Respekt x Doni 52 (R x Dn)

* In brackets are the abbreviations of the crosses used according to their parental forms names

Statistical analysis

The data on each measured index were summarized and averaged. The statistical deviation, the variation coefficient, the variation range and the statistical error were determined. The data were grouped according to crosses and progenies. Cluster analysis based on data for all investigated parameters was applied according to Rachovska et al. (2003). MS Office Excel, 2003 and 2013 were used to process the data and for variation analysis, and IMB SPSS Statistics 19 – for cluster analysis.

RESULTS AND DISCUSSION

The response of the individual hybrid combinations and the cultivars was rather diverse both as two separate groups and as individual genotypes within those groups (Table 3-7). Significantly lower variation of all indices in the hybrids was observed in comparison to the cultivars. Such behavior is related to the high theoretical uniformity of the first hybrid generation and to the typically higher plasticity of the cultivars (Acquaah, 2007). The data on the range of variation univocally indicated that although there was high variation in the cultivars, dramatic deviations in comparison to the hybrid combinations were not observed. This was an indication that the cultivars were stable under the specific conditions.

On the other hand, great deviations in the hybrids were not noticed, which was related to the lack of unfavorable inter-genomic interactions between the parental forms. In spite of these relations, the high differences in the variation both between the individual cultivars and between the individual hybrids implied considerable genetic diversity. This was emphasized also by the differences in the variation between the separate indices, since their variation coefficients did not follow a common tendency in each of the investigated genotypes. Similar data on the variation of each of the studied indices in cultivars and hybrids has been reported in triticale (Stoyanov and Baychev, 2015), as well as in other cereals (Stoyanov, 2013; Stoyanov, 2014a; Stoyanov, 2015).

In order to characterize the genetic diversity in the investigated genotypes, both the variation and the mean values of the individual indices are equally important. The differences observed in the two statistical indices with regard to each genotype allowed evaluation of the genetic diversity in the investigated plant sets not only in its absolute sense, bit also following some of its individual components. Therefore, each of the studied structural elements of yield was evaluated in details according to its variation.

Date to heading

Eight of the 11 investigated cultivars (with the exception of Respekt, Lovchanets and Blagovest) started heading on 12th May. The tendency toward occurrence of mass heading was constant, 6 out of 11 cultivars being in mass heading till 15th May, while Respekt, Lovchanets and Blagovest were late. The variation of this index was comparatively low, which could be related to the purposeful breeding for medium early genotypes.

Deviations from the initial tendency were observed in cultivar Atila, which had rather long and non-uniform heading, from 12th to 22nd May, with mass heading on 18th May. Such behavior was untypical for this cultivar, based on previous studies (Baychev, 2009). It is worth noting that heading was comparatively late according to other investigations on the same genotypes (Baychev, 2013). Such data are indisputably related to the effect of the environmental conditions. Different investigations on triticale (Motzo et al., 2001) and other cereals (Yan and Hunt, 2001; Stelmakh, 1993; Arnoldo Amaya et al., 1971; Pirasteh and Welsh, 1980) undoubtedly emphasize the role of the growing conditions for the date of heading. On the other hand, this index is very conservative as a tendency. This is underlined by the absence of a serious effect of the environment x genotype factor according to Motzo et al. (2001).

Table 3. Heading and height of plants from triticale cultivars and hybrids on nutrition area of 30/10 cm.

№ Cross	Variety / Cross	Date to heading, May				Plant height, cm			
		$\bar{X}\pm S_x$	VC, %	Range of variation		$\bar{X}\pm S_x$	VC, %	Range of variation	
12/1 4	At x Ak	14±0,2	8,19	12	15	124±1,2	5,23	116	139
13/14	At x R	17±0,4	9,08	14	20	123±2,2	6,86	112	140
24/14	Ak x B	15±0,2	6,68	12	14	121±1,5	7,01	97	134
25/14	Ak x I	15±0,2	5,97	14	17	122±1,9	6,78	98	141
26/14	Ak x Dr	13±0,4	8,6	12	15	127±4,4	9,75	102	144
28/14	Ak x Dn	14±0,2	6,71	12	16	121±1,3	6,07	103	133
34/14	R x B	15±0,2	6,46	14	18	114±1,4	4,91	117	129
35/14	R x I	15±0,2	7,16	14	17	123±1,9	7	103	142
36/14	R x Dr	14±0,8	15,16	12	18	116±3,6	8,22	104	131
38/14	R x Dn	15±0,3	10,44	13	20	122±1,8	7,64	103	137
1	Kolorit	15±0,3	13,58	12	18	110±1,1	6,43	96	122
2	Atila	18±0,4	14,61	12	22	116±1,6	8,47	87	130
3	Akord	16±0,2	9,91	12	20	117±1,3	8,16	85	137
4	Respekt	16±0,2	7,85	13	18	115±1,2	6,72	100	145
5	Bumerang	15±0,2	8,76	12	20	117±1,3	7,06	97	133
6	Irnik	15±0,2	8,89	12	18	110±1,2	6,35	95	122
7	Dobrudzhanets	15±0,3	11,93	12	19	109±1,6	9,01	87	128
8	Lovchanets	16±0,3	11,83	13	20	109±1,8	9,46	84	126
9	Doni 52	15±0,2	8,46	12	19	114±1,0	5,9	100	128
10	Blagovest	17±0,3	12,65	13	22	110±1,5	8,53	91	136
11	Borislav	14±0,1	6,44	12	16	111±0,6	3,57	103	118

A high number of the investigated crosses, 8 out of 10, also demonstrated lower variation in the date to heading. They, however, were characterized with rather variable dynamics of heading strictly dependent on the genotype. Therefore five crosses started heading on 12th May, one on 13th May, and four on 14th May. In contrast to the cultivars, the period of heading in the hybrid progeny was considerably narrower, with the exception of crosses 36/14, 38/14 and 13/14. Divergent data on heading in hybrids has been reported in other research, too, emphasizing the lack of a general regularity depending on the parental components used (Stoyanov and Baychev, 2015; Stoyanov and Baychev, 2016a). Due to the specific peculiarities of this index (Li et al., 2003), the investigated crosses had the possibility to combine heading periods of different duration from the parental forms. The different combinations allowed selection in the subsequent generations genotypes which possessed the target values of the breeding program. From this point of view, the studied hybrid combinations are a highly valuable source of genetic diversity according to the investigated index.

Plant height

The data on plant height are presented in Table 4. This index demonstrated very low variation values – between 4 and 9 %. Cultivars Atila, Dobrudzhanets and Lovchanets were with highest variation, while Borislav, Respekt and Doni 52 – with lowest, respectively. The means of the index did not vary widely in the respective cultivars. Akord, Respekt and Bumerang were with highest plant height, while Kolorit, Irnik Dobrudzhanets, Lovchanets and Blagovest – with lowest. The data on the variation of the investigated hybrids in the first generation were identical. Comparatively low percent of variation was observed – between 5 and 10 %. Crosses 26/14 and 36/14 were with the highest variation, while crosses 12/14 and 34/14 were with the lowest variation. The hybrid plants exceeded the means of the triticale cultivars with up to 10 cm, with the exception of cultivars 34/14 and 36/14, which remained comparatively shorter. The lack of significant variability in the height of the plants is a result of the purposeful selection carried out toward shorter plants (Baychev, 2013). The respectively higher plants in the hybrid combinations were related to a heterosis effect in this index (Stoyanov and Baychev, 2015).

Different investigations on the height of the plants from the cereal crops highlight the strict dependence of the index on the growing condition, and lower variation as a result of the interaction of the environment with the genotype (Guinta et al., 1999). This allowed searching for new combinations in the studied hybrid combinations in comparison to the parental forms. On the other hand, the very high and variable values of vegetative heterosis is reported in previous study on the same crosses (Stoyanov and Baychev, 2016a). As both in triticale (Herrmann, 2007; Oettler et al., 2001; Oettler et al., 2004) and in common winter wheat (Chwang et al., 1963) different investigations also reports similar heterosis values in the first hybrid generation, the direct differentiation of the crosses as sources of genetic diversity by this index is very limited. This was due to the presence of a clearly expressed tendency the plants to be higher than the parental forms. Furthermore, the variation in the heterosis expressions between the individual crosses implied different variation of the index in the subsequent generations, which is very valuable from a breeding point of view.

Spike length

The variation of the length of the spike in the investigated plants was low to moderate - 9.22-13.80% in the crosses, and 8.40-15.38% in the cultivars. This showed that the plants did not differ very much by the investigated index, as confirmed also by the means of the entire investigated plant set – between 13 and 15 cm. Such data are related to a higher level of uniformity typical of the first hybrid generation and the cultivars. The direction of breeding toward moderate length of spike is also emphasized. Cultivars Akord, Doni 52 and Borislav were characterized with the longest spikes, and cultivars Irnik, Kolorit and Blagovest – with the shortest. In the crosses, respectively, longest spikes were formed in 25/14, 26/14 and 38/14, and shortest - in 12/14, 13/14 and 34/14. In the investigated cultivars, the longer spikes could be usually referred to greater number of spikelets, without noticing a general tendency toward higher number of grains.

The length of the spike can be considered a characteristic which is influenced to a lower degree by the environmental conditions and which is related to a greater effect of the genetic factors (Stoyanov, 2013; Kashif and Khaliq, 2004). Nevertheless, the studies on cereals such as wheat (Fufa et al., 2005; Shahid, 2000; Mahmood and Shahid, 1993; Friend, 1965) have shown that spike length is directly connected to the general development of the plant, i.e. the spikes would be smaller if there is strong disturbance of the growth (under unfavorable factors). Such correlations are typical for stress effects active till the flowering stage since the length of the spike is already formed at that time (Stoyanov, 2014a). These peculiarities allow the assumption that strong stress did not occur at the initial phases of the development in the investigated plants because the different genotypes responded with similar values (Stoyanov and Baychev, 2016a). Therefore, low genetic variability may be assumed for this index. Such low diversity in the investigated cultivars and crosses is due to purposeful breeding toward a certain type of spike (Stoyanov and Baychev, 2016b). Although the values of this index are related to high variation in common winter wheat (Kashif and Khaliq, 2004; Fufa et al., 2005; Shahid, 2000; Mahmood and Shahid, 1993; Friend, 1965), a more stable nature is observed in triticale (Alheit et al., 2014; Baychev, 2013). The crossing of cultivars with similar spike length is respectively related to very low diversity in the progenies. This is confirmed by data reported in other investigations (Baychev, 1996).

Table 4. Length of spike and Number of spikelets in a spike in triticale cultivars and hybrids on nutrition area of 30/10 cm.

№ Cross	Variety / Cross	Length of spike, cm				Number of spikelets in a spike			
		$\bar{X} \pm S_x$	VC, %	Range of variation		$\bar{X} \pm S_x$	VC, %	Range of variation	
12/1 4	At x Ak	12,9±0,22	9,22	9,8	14,6	32±0,5	8,09	26	36
13/14	At x R	12,9±0,33	10,18	9,2	14,3	31±0,7	9,38	22	35
24/14	Ak x B	13,5±0,27	11,61	9,1	15,8	34±0,6	9,46	26	40
25/14	Ak x I	15,0±0,36	11,07	11,1	17,3	34±0,7	8,93	27	38
26/14	Ak x Dr	14,6±0,61	11,76	13,3	17,6	33±0,8	6,68	30	36
28/14	Ak x Dn	13,9±0,33	13,8	10,8	18,1	33±0,5	8,02	29	39
34/14	R x B	13,4±0,29	10,01	11,1	16,4	33±0,4	4,8	30	36
35/14	R x I	13,7±0,37	12,44	10,3	16,9	33±0,5	6,59	28	38
36/14	R x Dr	13,6±0,58	12,1	11,1	16,2	33±1,1	9,3	27	36
38/14	R x Dn	14,5±0,26	9,8	11	17,3	34±0,6	9,06	28	38
1	Kolorit	12,7±0,30	13,08	9,1	17,3	30±0,5	8,36	24	37
2	Atila	13,3±0,32	13,31	9,2	17,6	31±0,7	12,9	17	37
3	Akord	14,8±0,23	8,4	13,1	17,6	34±0,4	6,76	27	37
4	Respekt	13,5±0,24	9,61	10	16,1	34±0,4	6,21	30	37
5	Bumerang	13,6±0,24	9,83	11	17,3	33±0,4	6,92	28	38
6	Irnik	11,7±0,27	12,64	8,9	15,3	31±0,4	7,35	26	35
7	Dobrudzhanets	13,2±0,33	13,48	9,8	17,6	31±0,4	7,9	24	34
8	Lovchanets	13,3±0,37	15,38	8,5	17,6	30±0,6	10,45	21	35
9	Doni 52	14,2±0,33	12,81	8,7	17,6	33±0,6	9,46	23	38
10	Blagovest	12,4±0,38	16,72	8,9	16,6	32±0,6	10,08	26	39
11	Borislav	14,1±0,24	9,38	11,1	16,8	31±0,5	8,63	24	35

Number of spikelets per spike

The number of spikelets per spike in the investigated plants demonstrated low variation, similar to that of spike length. These low values, both in the cultivars and in the crosses, allow the assumption that this index, too, was little affected by the environment (Rawson, 1969). This was again confirmed by the small differences in the means of the index - 31-34 of the crosses and 30-34 of the varieties. Cultivars Akord, Respekt and crosses 24/14, 25/14 and 38/14 were with highest number of spikelets. Cultivars Kolorit and Lovchanets and crosses 12/14 and 13/14 were with lowest values.

The possibility that high level of stress affects this index was very low because its formation was completed during the booting stage (Rahman and Wilson, 1977). In this respect, a severe late frost with short action on the plants can have more significant effect (Warrick and Miller, 1995). Severe damages on the spike spindle, and hence – on the number of spikelets, are typical for plants which have been subjected to the effect of chemical agents in the early stages of treatment (colchicine, trifluralin) (Morrison et al., 1989). While the spike length follows the general development of the plant under conditions of drought or other stress, the number of spikelets remains unchanged (Friend, 1965; Rahman and Wilson, 1977). This is typical even for distant hybrids and amphoploids with seriously disturbed process of meiotic division (Stoyanov, 2015). This was the reason for the similar data observed in the investigated genotypes. Since the environment has low influence on the formation of the values due to the absence of strong stress effects, the differences determined both in the cultivars and in the crosses were to a high extent conditioned by genetic factors. Due to these characteristics, it can be claimed that the investigated genotypes are too similar by this index and are characterized with lower genetic diversity.

Number of grains per spike

In contrast to the preceding two indices, the number of grains per spike was characterized with very high variation in the investigated plants. Similar data are typical of such cereal crops as wheat, barley and triticale since the number of grains per spike is directly influenced by the environmental factors, which affect pollination and fertilization (Fisher, 1985).

In the cultivars and crosses, significant differences with regard to variation were not observed; it was within the range 16 – 25 % for the cultivars and 17 – 24 % for the crosses. Cultivars Blagovest, Boomerang, Kolorit and Irnik were with highest mean values, and Atila and Borislav – with lowest. These two cultivars had very high values of 1000 kernel weight, which is a character typical for varieties forming lower number of grains (Stoyanov and Baychev, 2015).

There is a significant tendency the forms with larger grain to form lower number of grains (Stoyanov and Baychev, 2015; Stoyanov and Baychev, 2016a). Among the crosses with highest values, 25/14 and 36/14 should be pointed out, and among those with lowest - 13/14 and 24/14. The differences between the means of the cultivars and crosses were not very big, which is related to the identical effect of the environment on the productivity. The variation of this index was very high under specific conditions of the environment, as well as under contrasting ones. An evidence for this are the data obtained by Guinta et al. (1999), Motzo et al. (2001), Ugarte et al. (2007), Lopez-Castaneda and Richards (1994), Fisher (1985). Such a tendency was not observed in the investigated crosses because there was no stress effect during heading and anthesis. This allowed a better differentiation of the genotypes since the fertility of the spikes depended on the genetic factors to a higher degree.

Table 5. Number of grains per spike and Grain weight per spike in triticale hybrids and cultivars on nutrition area of 30/10 cm.

№ Cross	Variety / Cross	Number of grains per spike				Grain weight per spike, g			
		$\bar{X} \pm S_x$	VC, %	Range of variation		$\bar{X} \pm S_x$	VC, %	Range of variation	
12/1 4	At x Ak	87±3,3	21	38	123	5,24±0,215	22,45	2,07	6,88
13/14	At x R	77±4,5	23,29	45	99	4,53±0,291	25,63	2,61	6,05
24/14	Ak x B	80±3,1	22,34	34	108	4,86±0,235	28,19	1,44	7,29
25/14	Ak x I	100±4,9	22,96	63	155	5,32±0,345	29,69	2,65	8,57
26/14	Ak x Dr	89±5,8	18,51	61	116	5,47±0,395	20,4	3,68	7,12
28/14	Ak x Dn	85±3,5	24,19	46	127	4,79±0,237	29,3	2,04	7,6
34/14	R x B	89±2,6	13,19	71	112	5,08±0,217	19,11	3,04	7,11
35/14	R x I	94±3,9	19,16	58	141	5,16±0,302	26,79	2,45	7,89
36/14	R x Dr	103±8,9	24,41	64	131	6,31±0,678	30,38	3,58	8,51
38/14	R x Dn	88±2,9	17,77	60	122	5,13±0,220	23,04	2,86	8,15
1	Kolorit	102±3,5	18,48	77	154	5,25±0,217	22,58	2,87	7,44
2	Atila	88±3,5	21,9	32	121	5,41±0,263	26,58	1,88	7,87
3	Akord	95±3,3	19,12	49	123	5,46±0,232	23,24	2,59	7,71
4	Respekt	93±3,5	20,86	67	155	5,11±0,265	28,39	3,24	9,91
5	Bumerang	104±3,1	16,29	79	140	6,08±0,224	20,17	3,5	8,98
6	Irnik	100±4,3	23,75	34	149	5,26±0,306	31,91	1,41	8,2
7	Dobrudzhanets	92±4,2	25,1	56	144	4,99±0,311	34,15	1,75	8,96
8	Lovchanets	99±4,6	25,16	36	141	5,21±0,278	29,25	1,9	8,28
9	Doni 52	97±4,2	23,67	36	133	5,06±0,268	29	1,25	7,56
10	Blagovest	107±4,7	23,98	64	166	5,74±0,298	28,42	3	9
11	Borislav	76±2,4	17,24	50	103	4,78±0,207	23,7	2,8	6,65

Since the number of spikelets was even less distinct, the identical data on the number of grains per spike indicated that pollination and fertilization, and the respective formation of number of grains per spike, occurred in similar manner even in different genotypes. Since these processes were characterized with higher susceptibility to the environmental conditions, it can be assumed that significant stress did not occur in the investigated plants during the anthesis stage. However, the high values of variation in this index relate on the other hand to the amphiploid nature of triticale and the impossibility for realization of higher number of grains (Baychev, 1990; Randhawa et al., 2015; Stoyanov and Baychev, 2016a; Sechnyak and Sulima, 1984). At the same time, the lack of significant stress on the plants and the comparatively high levels of variation imply significant genetic variability by this index. From a breeding point of view, such high genetic diversity allows positive selection and proper choice of initial material among the investigated cultivars.

Grain weight per spike

The weight of grains per spike as a weight index is characterized with a potential for high variation regardless of the investigated crop. This is due to the fact that the weight indices are most influenced by the environmental factors because they are related to multiple physiological processes (Stoyanov, 2013; Stoyanov and Baychev, 2015; Fisher and HilleRisLambers, 1978). Triticale is not an exception from this rule as evidenced by the data obtained from the investigated cultivars and crosses. The mean variation of the entire investigated plant set was 27 %, which implies a wide range of effect both of the genotype and the environment on the specific genotype. In the crosses, the variation was within 19 – 30 %, and in the cultivars – within 20-34 %. Such variations are indicative for the different expression of a certain genotype under the specific conditions of the environment. In this respect, cultivars Bumerang, Akord and Blagovest demonstrated highest values, while Dobrudzhanets, Doni 52 and Borislav had lowest values.

These data allowed assuming a very serious effect of the number of grains per spike on the formation of the weight indices during the specific period and its simultaneous considerable influence as a component of yield. Stoyanov and Baychev (2015) have proved the assumption while investigating the correlations in this crop for hybrid combinations. Such correlations have been reported in other investigations as well (Blanco et al., 2001; Dogan et al., 2009).

In the crosses, highest values were respectively observed in 25/14, 26/14 and 36/14, and lowest - in 13/14, 24/14 and 28/14. The considerably high differences formed in this index, within 1-2 g, showed that the spike productivity in the investigated forms was significantly different and was to a large extent related to the number of grains formed. It should be taken into account, however, that such relation was formed under favorable conditions of normal grains nutrition. Different investigations put emphasis on the fact that the high values of this index are related to suitable conditions for growing of the crop (Stoyanov and Baychev, 2015; Baychev, 2013; Guinta et al.,1999; Motzo et al., 2001; Ugarte et al., 2007). The possibility for expression of high values of the weight of grains per spike is, on the other hand, related to the possibility to better differentiate the investigated genotypes according to their productivity potential. The serious differences observed both in the group of the cultivars and in the group of the crosses emphasized the high genetic diversity in the expression of the trait. This firmly underlined the serious genetic diversity existing among the investigated genotypes.

1000 kernel weight

Certain differences were also observed between the investigated cultivars and crosses in connection with the effect of the genotype. Cultivars Atila and Borislav were with largest grains, and cultivars Kolorit and Irnik – with smallest. Among the crosses, 26/14 and 36/14 had the largest grains, and 25/14 and 35/14 – the smallest.

It was noted that the cultivars generated higher variation for this index, between 10 and 20 %, which was however significantly lower in the crosses – between 5 and 15 %. This was probably due to the high level of uniformity typical for the first hybrid generation. The differences in the investigated cultivars and crosses implied serious genetic diversity. It was due to the fact that although the characteristic was influenced by the environment under different levels of stress, it remained extremely conservative. Similar data have been reported for triticale and other cereals (Asif et al., 1999; Kashif and Khaliq, 2004; Stoyanov, 2013; Stoyanov and Baychev, 2015).

Thousand kernel weight is an index which varies within a wide range depending on the environment (Asif et al., 1999; Stoyanov, 2015). Certain research on common winter wheat point out that the stress factors have most significant effect on its formation regardless of their origin (Leilah and Al Khateeb, 2005). In this respect, a certain tendency occurred among the individual genotypes during a specific period of time related to heritability factors (Asif et al., 1999; Kashif and Khaliq, 2004). Therefore, regardless of the environment, the tendency between the genotypes remained the same within certain limits. This is so because the index characterizes the size of the grains. The proper nutrition of the grains is related to higher values of M1000. Its behavior has been confirmed in numerous investigations on triticale cultivars (Kutlu and Kinaci, 2010; Baychev, 2013) and common winter wheat forms (Leilah and Al Khateeb, 2005; Kashif and Khaliq, 2004). In the hybrid generations, however, different investigations (Stoyanov and Baychev, 2016a; Goral et al., 2015) have reported the presence of a high heterosis effect. This is the reason why the stress factors considerably complicated the reaction of the individual hybrid combinations. Therefore, the low values of a cross may be due to both genetic and stress factors. Since two of the investigated crosses combine parents with high and low M1000 values (12/14 and 13/14), such a response is to be expected in them.

In spite of the fact that the formation of the number of grains per spike occurred under the same conditions, and two of the crosses differed by the duration of their vegetative growth, the genetic factors in them had considerable effect. On the other hand, a large-grained genotype was not involved in cross 24/14 but in it, too, very high values of the index were determined. Such behavior of the investigated crosses considerably impedes the correct evaluation of the environmental effect and hence – the correct evaluation of the genetic diversity among the crosses.

Table 6. 1000 kernel weight in triticale hybrids and cultivars on nutrition area of 30/10 cm.

Cross No	Cultivar / Cross	1000 kernel weight, g			
		$\overline{X}\pm S_x$	VC, %	Range of variation	
12/1 4	Atila x Akord	60±0,9	7,87	43	67
13/14	Atila x respekt	59±1,4	9,28	48	66
24/14	Akord x Boomerang	60±1,3	12,55	42	73
25/14	Akord x Irnik	53±1,8	15,14	33	68
26/14	Akord x Dobrudzhanets	61±1,2	5,61	56	65
28/14	Akord x Doni 52	56±1,0	10,97	44	68
34/14	Respekt x Boomerang	57±1,7	13,66	43	70
35/14	Respekt x Irnik	54±1,7	14,32	31	65
36/14	Respekt x Dobrudzhanets	61±1,6	7,28	55	66
38/14	Respekt x Doni 52	58±1,0	9,29	48	67
1	Kolorit	51±1,6	16,79	33	69
2	Atila	61±0,4	12,42	48	80
3	Akord	57±0,3	12,68	41	69
4	Respekt	55±0,2	11,53	43	65
5	Boomerang	58±0,1	10,71	44	71
6	Irnik	52±0,6	16,87	32	68
7	Dobrudzhanets	54±2,0	20,21	30	78
8	Lovchanets	52±1,0	10,1	40	59
9	Doni 52	52±1,3	13,69	30	64
10	Blagovest	53±1,2	12,49	39	66
11	Borislav	62±1,6	13,69	44	74

Spike density

Length of spike and number of spikelets per spike, being indices less influenced by the environment, allow following and more thoroughly characterizing certain genotypic factors (Stoyanov, 2014a). The relationship between them, which accounts for the density of the spike, is an important breeding characteristic showing the efficiency of the productivity of a certain type of spikes (Stoyanov, 2014b). This allows evaluating the degree of efficiency of the long spikes from a breeding point of view. This index was marked by very low variation, similar to TKW. In the investigated cultivars and crosses, the variation was low – 5-13%

and 4-10%, respectively. Such values indicate that this index is resistant to certain effects of the environment and varies within a very narrow range. In the tested plant set, the mean value was 2.4 spikelets per linear cm. It is worth mentioning that the investigated crosses differed very little in this respect; in many cases the differences may be due to and can be explained with statistical error. It can be pointed out in this relation that the lowest values from the crosses were formed in 12/14, and the highest – in 24/14.

In the cultivars, respectively, the highest values were that of Irnik, and the lowest – of Borislav. These data allow the assumption that the greater number of spikelets per length unit is related to the formation of smaller and more numerous grains. This was emphasized through the values of NGS and TKW of cultivars Bumerang, Irnik and Blagovest, while the opposite tendency was evident in cultivar Borislav. The similarity of the crosses to the cultivars in the intermediate group (approximating value 2.4) also highlighted the same tendency.

Stoyanov and Baychev (2015) have reported similar data on direct and reciprocal crosses of triticale. Therefore, it can be pointed out that this index gives indirect information on the number of grains per spike. Based on the above relations, it can be concluded that in the more dense spikes the grain size is determined by the availability of sufficient space between the spikelets. The investigated crosses formed spikes, which can be described as balanced in structure.

Previous investigations have stressed the significance of this trait as being very stable (Stoyanov, 2014a). In this respect, the absence of variability by this index is related to the identical structure of the spikes of the investigated genotypes and the low genetic diversity among them.

Fertility

As in a typical cereal crop, the anthesis of triticale is strongly influenced by the conditions of the environment (Sechnyak and Sulima, 1984). Therefore, triticale fertility can be regarded as strongly affected by both the environmental conditions and the genetic factors (Giunta et al., 1993), similar to the index M1000.

A comparatively high fertility for this crop was observed in the investigated crosses and cultivars – averagely 2.9 grains per spikelet. On the one hand, this was due to the larger nutrition area, and on the other – to the favorable conditions of the environment for pollination and formation of grains (Fischer, 1985). Although this index is basically related to the number of grains per spike, its relation to the weight indices should not be ignored; regardless of the occurrence of pollination and fertilization, the subsequent grain formation is significant for this index. In this respect, the high dependency of the index on the weight of grains per spike should be emphasized.

Table 7. Spike density and Fertility of triticale cultivars and hybrids on nutrition area of 30/10 cm.

№ Cross	Variety / Cross	Spike density				Fertility of spikelet			
		$\bar{X}$±S_x	VC, %	Range of variation		$\bar{X}$±S_x	VC, %	Range of variation	
12/1 4	At x Ak	2,5±0,03	5,73	2,3	2,8	2,7±0,09	18,93	1,5	3,7
13/14	At x R	2,4±0,03	5,1	2,2	2,7	2,5±0,13	20,85	1,5	3,2
24/14	Ak x B	2,5±0,04	8,23	2,2	3,4	2,3±0,08	18,99	1,3	3,5
25/14	Ak x I	2,3±0,02	4,82	2,1	2,6	2,9±0,11	18,26	1,8	4,2
26/14	Ak x Dr	2,3±0,08	10,22	1,9	2,7	2,7±0,16	17,03	1,9	3,2
28/14	Ak x Dn	2,4±0,03	7,94	2,1	2,8	2,5±0,08	18,59	1,5	3,3
34/14	R x B	2,5±0,05	8,63	2,2	3	2,7±0,06	10,43	2,1	3,2
35/14	R x I	2,5±0,06	10,35	2,2	3,3	2,7±0,09	14,66	1,8	3,7
36/14	R x Dr	2,4±0,04	4,45	2,2	2,6	3,1±0,20	17,65	2,2	3,9
38/14	R x Dn	2,4±0,03	7,09	2	2,6	2,6±0,08	15,89	1,8	3,4
1	Kolorit	2,4±0,05	11,12	2	3,1	3,4±0,10	16,06	2,3	4,7
2	Atila	2,4±0,05	11,15	1,9	3	2,8±0,09	18,18	1,3	3,4
3	Akord	2,3±0,03	7,62	2	2,7	2,8±0,11	20,87	1,5	4,5
4	Respekt	2,5±0,03	7,51	2,2	3,1	2,7±0,09	17,82	2	4,3
5	Bumerang	2,5±0,04	8,96	2,1	3,1	3,1±0,09	15,25	2,3	4,1
6	Irnik	2,7±0,05	10,53	2,2	3,6	3,2±0,13	22,03	1,3	4,5
7	Dobrudzhanets	2,3±0,05	10,71	1,8	3	3,0±0,12	21,69	1,8	4,2
8	Lovchanets	2,3±0,04	9,59	1,9	3,2	3,3±0,12	19,86	1,7	4,3
9	Doni 52	2,4±0,04	8,3	2	2,8	2,9±0,11	20,48	1,6	3,9
10	Blagovest	2,6±0,06	13,28	2	3,3	3,3±0,11	17,72	2	4,5
11	Borislav	2,2±0,02	5,42	2	2,5	2,4±0,06	13,16	1,9	3

Among the investigated cultivars, Kolorit, Lovchanets and Blagovest formed highest fertility, and Borislav and Respekt – lowest. Among the investigated crosses, the fertility of 36/14 and 25/14 was highest, and that of 25/14 and 28/14 – lowest. It is worth mentioning that most of the crosses formed fertility at the level of the low fertility of the cultivars. It is also necessary to emphasize that regardless of these data the first hybrid generation realized fertility above 2 grains per spikelet, which is especially important in the breeding of triticale (Baychev, 1990).

The low values observed in the crosses are indicative of the serious interactions, which occurred at the level of the first hybrid generation. This is due to the delicate balance between the wheat and the rye subgenome in the cultivars, which is missing in the hybrid plants (Lelley, 2006). Such dependencies are the reason an extremely high genetic diversity to be realized in the subsequent generations.

The data on the variation of the investigated cultivars and crosses unanimously showed that the genotypes differed considerably by both their productivity and the range of their reaction to the specific growing conditions. This is indicative for the considerable genetic diversity existing in triticale, even according to the small number of investigated genotypes. This diversity was confirmed also by the cluster analysis carried out (Figure 1). Each cluster was formed on the basis of the studied structural elements of the productivity of the studied genotypes. This allowed grouping the individual genotypes by their complex phenotypic expression at the low level of stress of under which the cultivars and the crosses were grown.

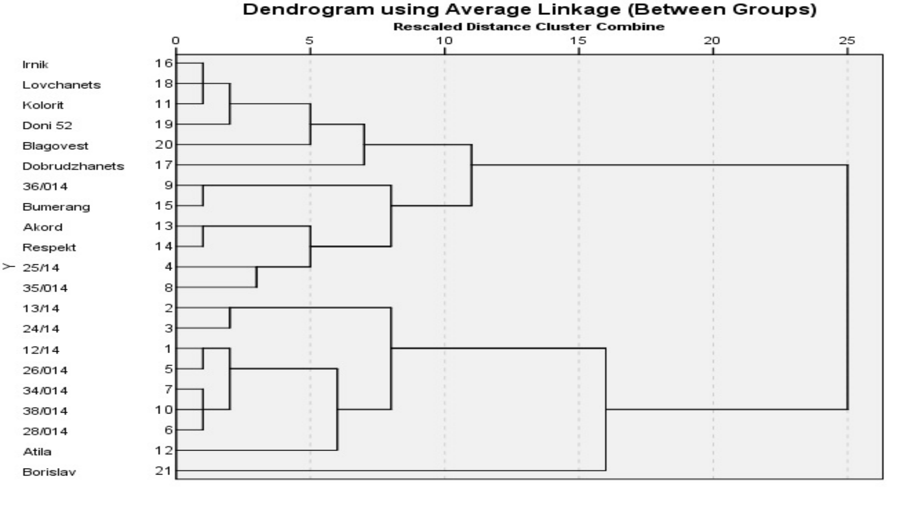

Fig. 1. Dendrogram of a complex cluster analysis by structural elements of yields in triticale cultivars and crosses

Distinct clusters were outlined, in which either the cultivars or the crosses were predominant. On the other hand, it was evident that neither of the two large clusters was homogenous (they contained predominantly cultivars or crosses) but was characterized with high genetic variability. This was emphasized by the various Euclidian distances formed between the separate genotypes. On the other hand, a part of the crosses were grouped together with the cultivars and vice versa. Such lack of homogeneity and presence of specific clusters make the crosses significant enrichment to the genetic diversity. Simultaneously, the close grouping of cultivars such as Atila and Borislav with the hybrid combinations was indicative for the different ways of the formation of their productivity in comparison to the other cultivars. These data have been confirmed by previous studies (Stoyanov and Baychev, 2015).

On the other hand, a grouping of hybrid combinations in close proximity to the cultivars is related to their high productivity, and also to a higher plasticity. These crosses (35/14, 36/14), together with crosses 28/14 and 38/14, were characterized with the highest variation coefficients for most of the investigated indices. Such high level of variation implies considerable diversity in the response to the environmental conditions in the next hybrid generations. Therefore, it can be assumed that the crosses are a source of significant genetic variability, which makes them the most valuable breeding material for improvement of triticale.

CONCLUSIONS

The investigated triticale cultivars were characterized with greater variation of the investigated indices in comparison to the hybrid combinations, which can be related to the high homogeneity of the first hybrid generation and the high plasticity of the cultivars to the environmental conditions. The cultivars and the crosses demonstrated highest variation by the indices number of grains per spike and grain weight per spike; this highlighted their wide range of response to the environment regardless of their genetic uniformity. In parallel, the different values of the specific genotypes allow the assumption that a considerable genetic diversity exists in the investigated plant set with regard to spike productivity. Crosses 28/14, 35/14, 36/14 and 38/14 had highest values and highest variation, which makes them valuable breeding material for development of genetic variability and for improvement of winter hexaploid triticale.

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