Evaluation of Morphological Diversity, Mitotic Instability and Fertility in Tritipyrum

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Abstract

Tritipyrum is prone to mitotic instability, stiff straw and low fertility. A complete randomized design was used to evaluate morphological diversity, fertility and grain yield of 7 new synthetic Tritipyrum lines and their F_1 offspring'sin a cross with bread wheat. Fertility, harvest index, biological yield, 1000-grains weight (TGW) and grain yield of Tritipyrum derived genotypes were significantly lower than bread wheat and Triticale. The morphology of Tritipyrum was found to be similar with wheat lines and hybridization between Tritipyrum and bread wheat was efficient for developing early maturelines and reducing stiff straw and plant height. Results showed that mitotic instability in small size grains (TGW< 30 g) were significantly higher than heavy grains (TGW> 30 g) in both parental and F_2progenies .Euploidyof Tritipyrum derived genotypes were significantly lower than Triticale and bread wheat. Results suggested that the larger and heavier grains show higher frequency of euploidy and that can be useful for maintenance of the genetic stability of Tritipyrum genotypes. Tritipyrum as a new man made cereal is a good genetic resource for wheat breeding and may complements the role of bread wheat wherever environmental condition is not suitable for wheat cultivation.

Keywords: Amphiploid, Aneuploid, Euploid, Stiff Straw, Tritipyrum

Introduction

The incorporation of alien genetic material from *Thinopyrum* into common wheat through wide hybridization has increased the genetic diversity for improvement of drought and salinity tolerance, disease resistance and others traits in wheat (Chen Q, 2005; Allahdoo, et al., 2009). A new cereal, Tritipyrum, with a range of amphiploidslevels between *Triticum spp*. and *Thinopyrum spp*. offers such a new chance. HexaploidTritipyrum (2n=6x=42, $AABBE^bE^b$) derived from Triticum durum (2n=4x=28, AABB) and *Thinopyrumbessarabicum* (2n=2x=14, E^bE^b) has the potential of becoming a new high salt tolerant cereal crop (King, et al., 1997; Shahsavand hassani, 1998; Shahsavand hassani, et al., 1998; Shahsavand hassani, et al., 2006; Allahoda, et al., 2009).

Tritipyrum, due to low fertility and morphological problems, will not fulfill the requirements of a successful commercial salt tolerant crop. Tritipyrum is prone to problems which had been observed in early Triticales, e.g. chromosome instability and low fertility (Shahsavand hassani et al., 1998; Shahsavand hassani, et al., 2000; Shahriari, et al., 2012).Tritipyrum exhibited a considerable amount of instability in the form of aneuploidy with a proportion of plants having lost chromosomes and some having addition chromosomes (Shahsavand hassani, et al., 2000).

The assessment of today's Tritipyrum situation in comparison with Triticale and the possible problems facing breeders in future is not an easy undertaking.Research works on Triticale in recent years show the importance of this first man-made cereal (Oudjehih and Boukaboub, 2000; Tosun, et al., 2003; Akgun and Altindal, 2010). There is no reason that the same success should not be achieved in the case of Tritipyrum (Shahsavand hassani, et al., 2000; Kamyab, et al., 2012, Shahriari, et al., 2012). Therefore, given the importance of new man made crops, the aim of this study were to study the morphological diversity and fertility of Tritipyrum and to evaluate its mitotic instability as compared with triticale and bread wheat.

Materials and Methods

Tritipyrum primary lines including Az/b; La/b; (Ka/b)(Cr/b),F_5; (Ma/b)(Cr/b); (St/b)(Cr/b),F_3; Ma/b and Cr/b were selected, based on results of a previous study that showthese genotypes were the most promising lines for growing conditions in Iran (Shahsavand hassani et al., 2000). Tritipyrum lines were crossed with a bread wheat cultivar named Navid. Spikes of each line of Tritipyrum(2n=6x=42, $AABBE^bE^b$) were emasculated, which subsequently pollinated with pollen grains from Navid and their F_1 and F_2 seeds were produced. SevenTritipyrum genotypes along with their F_1 progenies, Triticale 4118 promising line and the cultivar Navidwere sown in a greenhouse based on a complete randomized design scheme with 3 replicates. The seeds of Triticale and bread wheat collected from the research farm of College of Agriculture, Shiraz University, Iran. Total tiller and fertile tiller number, day to flowering and maturity, plant height at maturity, stiff straw (%of spike shattering), grain yield per plant, 1000-grains weight, grain number per spike, biological yield, harvest index and fertility was recorded for all genotypes. Fertility was quantifiedbased on the number of grain per spikelet of the main spike (Elneskog Staam and Marker, 2002). For mitotic chromosome counting, the primary Tritipyrum lines and their F_2 progenies were evaluated in a complete randomized design. The Tritipyrum lines and the promising Triticale line were divided into two classes based on their 1000-grains weight (less and higher than 30 g). "Navid" bread wheat cultivar was classified as the heavy grain class (heavier than 30 g). Grains were germinated in Petri dish and then planted in small pots that were allowed to grow to form a proper size for chromosome counting. The chromosome number of all lines was counted in 5 metaphase plates by minor modification of the conventional root tip squash protocol (Tosun, 1999; Oudjehih and Boukaboub, 2000; Oudjehih and Bentouati, 2006). All data were subjected to the analysis of variance using SPSS statistical software. Means were compared using Duncan multiple range test (DMRT) at the 0.05 significance level. The Ward hierarchical cluster analysis (Jaynes, et al., 2003) wasperformed using the means of grain yield, fertility and TGWfor all genotypes. Pearson correlation coefficientswere also calculated (Assad, 1997).

Results and Discussion

Yield components of hexaploidTritipyrum-derived genotypes and the promising Triticale line (no. 4118) and bread wheat “Navid cultivar” are evaluated(Table 1). Grain yield ($p < 0.05$) and TGW ($p < 0.01$) of Tritipyrum derived genotype was significantly lower than Triticale and bread wheat. Fertility of 5 parental Tritipyrumgenotypes had no significant differences with Triticale ($p > 0.05$). The fertility of St/b*Cr/b (F_3) Tritipyrum genotype was similar to bread wheat and Triticale. Harvest index and biological yield of Tritipyrum derived genotypes was significantly lower than that of Triticale and bread wheat. The overall results demonstrated that Tritipyrum have reasonable agronomical performance and moderate fertility which is in agree withKing, et al., (1997), Shahsavand hassani, et al., (1998, 2000, 2006) and Shahriari, et al., (2012).

Analysis of variance showed significant differences among Tritipyrum derived genotypes, bread wheat and Triticale for height, day to flowering, day to maturity and stiff straw (Table2). Plant height variations showed that Tritipyrum derived genotypes were significantly taller than “Navid” semi dwarf bread wheat. There is no significant difference between plant height of some of Tritipyrum derived genotypes and that of Triticale. There was no significant difference between tiller and fertile tiller number of Tritipyrum derived genotypes and that of bread wheat and Triticale. All parental Tritipyrum genotypes were significantly late flowered and late matured than bread wheat and Triticale. There was no significant difference between maturity of four Tritipyrum/bread wheat F_1[Ma/b*Navid(F_1), St/b*Cr/b (F_3)*Navid(F_1), Ma/b*Cr/b*Navid(F_1) and Ka/b*Cr/b(F_5)*Navid(F_1)] and their parental bread wheat “ Navid cultivar”. Tritipyrum derived genotypes stiff straw was significantly more than that of bread wheat and Triticale. Tritipyrum/wheat F_1 offspring’s showed significantly lower stiff straw than their parental Tritipyrum lines. In general, the morphology of Tritipyrum derived genotypes was predominantly similar to “Navid” Iranian bread wheat (King et al., 1997). The experiment showed that hybridization of bread wheat and Tritipyrum is hopeful for reducing plant height, stiff straw and for early maturityimprovement.

Analysis of variance indicated that the effect of grain weight on mitotic chromosome stability was significant (Table 3). There was a significant difference ($p<0.01$) between euploid plants means within light and heavy grains of Tritipyrum derived genotypes and Triticale. Chromosome number investigation of the light-grain class showed that Tritipyrum-derived genotypes were significantly more mitotically instable than Triticale and four F_2 progenies showed significantly high mitotic instability than their parental Tritipyrum genotypes. Euploid seedlings means among heavy grains of all Tritipyrum-derived genotypes (parental Tritipyrum genotypes and F_2 progenies) were significantly lower than Triticale and bread wheat as control. Euploied seedlings means among heavy grains of F_2 progenies showed no significant difference with their parental Tritipyrum genotypes. Chromosome counting of hexaploidTritipyrum-derived genotypes (42chromosomes) exhibited a considerable (12- 32.1 %) instability in the form of aneuploidy. The results were in agreement with King et al., (1997), Shahsavand hassani, et al., (1997), Shahsavand hassani, et al., (2000), Siahsar, et al., (2011), and Shahriari, et al., 2012. It has been suggested that the screening of larger and heavier grains could give a higher frequency of euploids and that grain weight was significantly affected by mitotic chromosome stability in segregation generations. Similar investigations in other crops displayed a close relationship between grain characteristics and aneuploid frequency (Tsuchiya, 1973; Suarez and Favret, 1986; Oudjehih and Boukaboub, 2000; Tosun, et al., 2003;Akgun and Altindal, 2010; Shahriari, et al., 2012).

Table 1. Yield components of hexaploidTritipyrum-derived genotypes compared with a promising Triticale (4118) and bread wheat “Navid cultivar”.

Genotypes	Line/cultivar/progenies	grain yield (gr/ plant)	Fertility (grain number / spikelet)	1000-grains weight (gr)	grain no/spike	biological yield (gr/plant)	harvest index
Primary Tritipyrum lines	(Ka/b)(Cr/b), F_5	2.93c	2.86d	32.87d	35.77cde	11.54cde	25.43e
	La/b	3.07bc	3.07bcd	33.7cd	34.56f	11.43def	26.81cde
	Az/b	3.03bc	2.9dc	33.74cd	35.07def	11.44def	26.52cde
	(Ma/b)(Cr/b)	3.13bc	3.03bcd	34.33cd	34.93ef	11.93b	26.26de
	(St/b)(Cr/b), F_3	2.98c	3.5a	33.9cd	35.5cdef	11.76bcd	25.49e
	Ma/b	2.56d	3.07bcd	33.1d	36.17bc	11.63bcde	22f
	Cr/b	2.97c	3.26abc	34.74cd	35.97bcd	11.4def	26.31de
Triticale	4118	4.03a	3.4ab	45a	36.9a	12.47a	32.35ab
Bread wheat	Navid	4.23a	3.6a	41.5b	37.33a	12.8a	33.08a
Tritipyrum/bread wheat(F_1)	(Ka/b)(Cr/b),F_5/Navid (F_1)	3.27bc	2.1e	34.7cd	33.01g	11.24ef	29.06cd
	(La/b)/Navid(F_1)	3.4b	1.83ef	34.71cd	32.09g	11.46def	29.65bc
	(Az/b)/Navid(F_1)	3.1bc	2.07ef	33.6cd	32.5g	11.12f	27.9cde
	(Ma/b)(Cr/b)/Navid(F_1)	3.23bc	2ef	33.97cd	32g	11.5def	28.12cde
	(St/b)(Cr/b), F_3/Navid(F_1)	3.26bc	1.73f	33.57cd	31.01h	11.9bc	27.45cde
	(Ma/b)/Navid(F_1)	3.03bc	2.04ef	35.26c	31.9gh	11.43def	26.53cde
	(Cr/b)/Navid(F_1)	3.25bc	1.97ef	35.23c	32.3g	11.6bcde	28.16cde
	F test	2.6*	2.45*	2.53*	4.5**	4.64**	5.1**
	CV%	8.7	9.02	10.23	8.89	7.987	8.54

Columns with the same letter(s) were not significantly different based on Duncan test at p≤0.05. * and ** significant at 0.05 and 0.01, respectively.

Table 2. Morphological diversity and stiff straw(%) of Tritipyrum derived genotypes compared with Iranian Bread wheat and a promising Triticale (4118).

Genotypes	Line/cultivar/progenies	Day to Flowering	Days to Maturity	Tiller NO	Fertile Tiller NO	Stiff Straw (%)	Plant height (cm)
Tritipyrum parental genotypes	(Ka/b)(Cr/b), F_5	149.33ab	181.67a	7.29b	5.7b	25a	74.67hi
	La/b	152a	180.34abc	9ab	5.81b	26.3a	139.33a
	Az/b	146e	182.66a	8.31ad	6.7ab	25.42a	109.6b
	(Ma/b)(Cr/b)	145.67e	180.64ab	8ab	6ab	25.2a	98.3d
	(St/b)(Cr/b), F_3	148.33cd	182a	8.25ab	6.3ab	23a	90.7e
	Ma/b	146.67ed	182.54a	4.9ab	7.3ab	24.7a	76gh
	Cr/b	151ab	181.41a	9ab	7.35ab	24.66a	74.5hi
Triticale	4118	131j	170g	9.7a	8.29a	3.2c	76.2gh
Bread wheat	Navid	135.32j	174f	9ab	7.29ab	6c	64.4k
Tritipyrum*bread wheat F_1 progenies	(Ka/b)(Cr/b),F_5/Navid (F_1)	141hg	176def	5ab	5.4b	13.4b	72ij
	(La/b)/Navid(F_1)	142.33fgh	177.7de	8ab	6.39ab	12.3b	102.7c
	(Az/b)/Navid(F_1)	143.33f	178.65bcd	8.1ab	6.5ab	14.5b	82.29f
	(Ma/b)(Cr/b)/Navid(F_1)	143fg	175.53ef	8.67ab	7ab	14b	87.71g
	(St/b)(Cr/b), F_3/Navid(F_1)	140.33h	176.45def	8.33ab	6.7ab	14b	77gh
	(Ma/b)/Navid(F_1)	141.67fgh	174.21f	7.67ab	6b	15b	70.65j
	(Cr/b)/Navid(F_1)	141.31fgh	178cde	8.2ab	7ab	13.1b	70.4j
	F test	8.04**	9.02**	1.03ns	0.95ns	7.9**	7.8**
	CV%	8.5	7.2	7.9	6.4	9.1	8.3

Columns with the same letter(s) were not significantly different according to Duncan test at p≤0.05. * and ** significant at 0.05 and 0.01, respectively.

Table 3. Mean comparison between euploid plants in light grain and heavy grain genotypes(60 seedlings per replication)..

Genotypes	Line/cultivar/progenies	Heavy grains Euploid seedling Number (2n=42)	Heavy grain aneplody (%)	Light grains Euploid seedling Number (2n=42)	Light Grains aneoploidy (%)	Mean differences (LSD)
Primary Tritipyrum lines	(Ka/b)(Cr/b), F_5	47.3def	21.2	44.67 bc	25.6	2.63**
	La/b	49.5bcde	17.5	46.3 b	22.8	3.2**
	Az/b	51bcd	15.0	46.7 b	22.2	4.3**
	(Ma/b)(Cr/b)	49bcde	18.3	45 bc	25.0	4**
	(St/b)(Cr/b), F_3	50bcde	16.7	46.3 b	22.8	3.7**
	Ma/b	51.67bc	14.0	45.6 b	24.0	6.07**
	Cr/b	52.7b	12.2	46.7 b	22.2	6**
Triticale	4118	58a	5.0	54 a	5.0	4**
Bread wheat	Navid	60a	0.0	-	-	-
Tritipyrum/bread wheat(F_2)	(Ka/b)(Cr/b),F_5/Navid (F_2)	45f	25.0	40.7 e	32.2	4.3**
	(La/b)/Navid(F_2)	47.3def	21.2	42.3cde	29.5	5**
	(Az/b)/Navid(F_2)	48.3cdef	19.5	41de	31.7	7.3**
	(Ma/b)(Cr/b)/Navid(F_2)	47ef	21.7	42cde	30.0	5**
	(St/b)(Cr/b), F_3/Navid(F_2)	48.6cdef	19.0	41de	31.7	7.6**
	(Ma/b)/Navid(F_2)	50.3bcde	16.3	42.3cde	29.5	8**
	(Cr/b)/Navid(F_2)	49.3bcde	17.8	44bcd	26.7	5.3**
	F test	8.5**	-	9.01**	-	-

Columns with the same letter(s) were not significantly different according to Duncan test at p≤0.05. * and ** significant at 0.05 and 0.01, respectively.

The cluster analysis technique is used for parental selection in breeding programs (El-Deeb and Mohamed, 1999; Leilah and Al-Khateeb, 2005) and/or crop modeling (Jaynes, et al., 2003). Squared Euclidean distance was able to cluster genotypes using the Ward hierarchical cluster analysis based on grain yield, 1000 grains weight and fertility (Figure 1). All genotypes separated into two main groups including 1) Tritipyrum derived genotypes and 2) Triticale and bread wheat. Cluster analysis showed that Triticale is more resemble than Tritipyrum to bread wheat. These data emphasized that more breeding researchesare neededto overcome limitations against Tritipyrumcultivations.

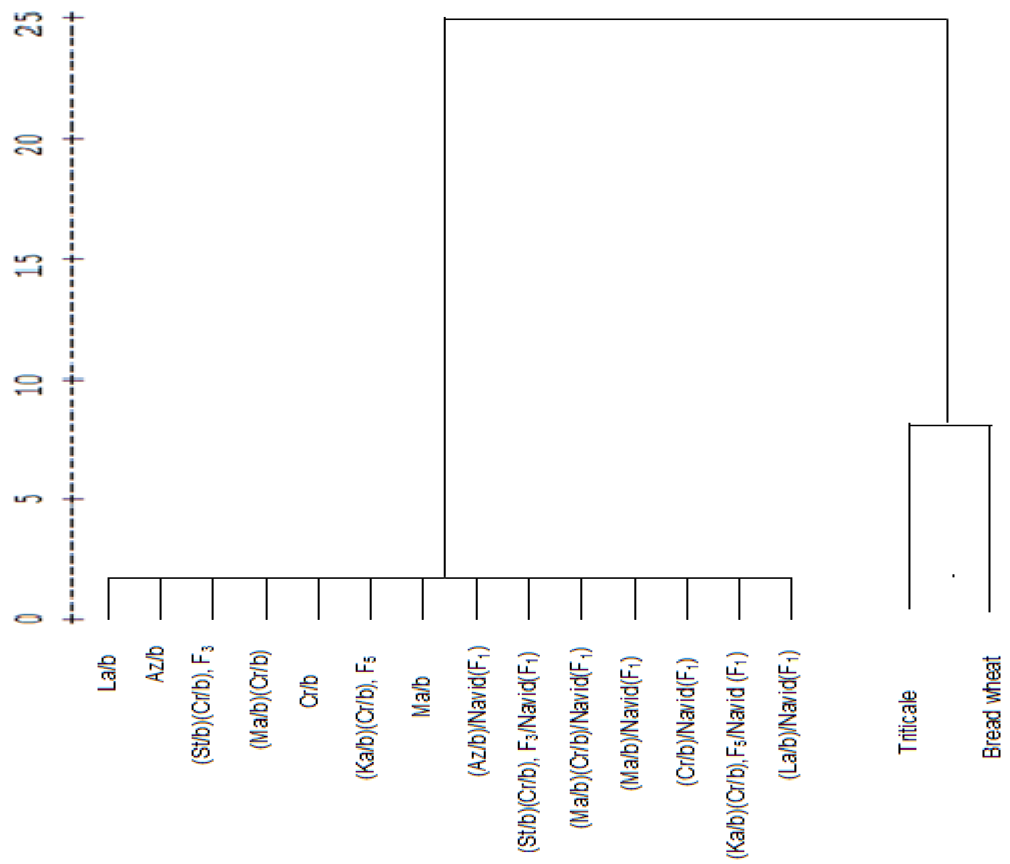

Figure 1. The tree dendrogram of cluster analysis (Squared Euclidean distance) for grain yield, fertility and 1000 grains weight in Tritipyrum derived genotypes, promising Triticale line and Iranian bread wheat cultivar ‘Navid’.

Conclusion

Tritipyrum as a new man made cereal may complement the role of bread wheat in regions in which environmental conditions are not suitable for wheat cultivation and is a good genetic resource for wheat breeding, but more breeding researches are needed to overcome its mitotic instability, low fertility and stiff straw by producing the secondary Tritipyrum genotypes via hybridization the primary lines with bread wheat cultivar.

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