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Meiosis in hexaploid triticale

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ABSTRACT

Meiosis was studied in five high-yielding and five low-yielding triticale genotypes. The percentage of metaphase I cells without univalents was 75.0%. The average number of univalents per cell was 0.56. Percentage of regular anaphase I cells was 80.1% and the average number of laggards per anaphase I cell was 0.48. On the other hand, 77.1% of anaphase II cells had regular chromosome distributions and the average number of laggards per anaphase II cell was 0.64. The percentage of tetrads without micronuclei was 73.5%, and the mean number of micronuclei per tetrad was calculated as 0.77. The percentage of pollen viability was 74.5%. It was determined that high-yielding genotypes were usually better than low-yielding genotypes in all traits.

INTRODUCTION

The main purpose of synthesizing triticale is to combine the grain yield and quality of wheat with the vigor and hardiness of rye. Many natural barriers interfere with the development of functional new species from wide crosses. Among the major hurdles in the development of triticale were cross-incompatibility, embryo starvation, hybrid necrosis, sterility, cytogenetic malfunction, and associated endosperm malformation. Ways were developed to overcome and avoid most of the barriers and thus improve high-grain-yield triticales (Zillinsky, 1985). However, problems such as meiotic irregularities, aneuploidy, and sterility were not fully overcome.

Triticale has combined genomes of two different genera and thus shows meiotic irregularities, sterility, and low grain yield. Obtaining results on meiosis varied with genotypes (Hafiz and Larik, 1984; Gupta and Priyadarshan, 1987; Chaubey and Khanna, 1988; Manero de Zumelzu et al., 1992).

This work differs from many other studies on meiosis in triticale because the materials were selected on the basis of grain yield (high and low yields). The goals of this study were (i) to determine the meiotic characteristics of different triticale genotypes and (ii) to compare high- and low-yielding triticale genotypes based on meiotic characteristics.

MATERIAL AND METHODS

In this study, the high-grain-yielding triticale genotypes used were Nutria 7272 (146 kg/da), Iguana 4–2 (141 kg/da), Hare 2 (121 kg/da), Bgl (118 kg/da), and Merino (118 kg/da)). Low-grain-yielding triticale genotypes were Hare 7265 (83 kg/da), Trasca 87 (79 kg/da), Hare 263 (78 kg/da), Alamos 83 (64 kg/da), and Hare 212 (54 kg/da) (Akgün et al., 1997).

The experiment was carried out in a completely randomized design with three replications. One replication contained five pots from each genotype (total 50 pots). In each genotype, five seeds were sown into each pot containing a mixture of soil: farm manure: sand (1:1:1) (total 15 pots from each genotype). Two seedlings were removed and the remaining three seedlings were grown under greenhouse conditions.

The correlations between grain yields, metaphase I univalents, anaphase I and II laggards, micronuclei per tetrads and pollen viability were calculated using Statistica for Windows program (Statsoft Inc., 2001).

Investigation of meiosis

The procedure of Sağsöz et al. (2002) was used for fixation. Spica were collected from fifty plants from

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each genotype, one spike from each plant. Spica were fixed in Carnoy's solution for 48 h, then 70% ethanol was added to the tubes and they were refrigerated.

Meiotic preparations were done with the aceto orcein squash technique (Moore, 1963). Each spike included (a) univalents in metaphase I, (b) anaphase I separations, laggards, and bridges, (c) regular and irregular separations in anaphase II, and (d) a number of micronuclei in tetrads. Meiotic investigations were carried out on euploids (2n = 42). Euploids were determined by chromosome counting in anaphase I.

Pollen viability

Pollen viability was determined from fifteen plants that were studied for meiosis in each genotype. Pollens were stained in 1% safranin for 1 h. Dark-stained pollens are viable and light-stained pollens are unviable (Sağsöz et. al., 1997).

RESULTS AND DISCUSSION

Metaphase I (MI)

Table 1 presents the metaphase I cells with and without univalents and mean number of univalents per cell. The variation between genotypes in the percentage of metaphase I cells without univalents and mean number of univalents per cell were significant. In one high-yielding genotype (Nutria 7272), the frequency of cells with univalents and the number of univalents per cell were lower than in four low-yielding genotypes. Other high-yielding genotypes did not differ from the low-yielding ones. Among metaphase I cells, 75.0% had no univalent. Nutria 7272 had the highest frequency of MI cells without univalents (81.4%), and Hare 212 had the lowest frequency (62.7%).

The number of univalents is a good indicator of meiotic stability in triticale, which was 0.56 and varied from genotype to genotype. Nutria 7272, which had the most regular MI cells, had the lowest number of univalents per cell (0.39), while Hare 212 had the highest number of univalents per cell (0.90). In the MI cells with univalents, the frequency of cells with two univalents was higher than others, and followed by the cells with 1, 4, 3, 5, 6, and 7 univalents, respectively.

Univalents of 2 or 4 in the cells may originate from desynapsis of bivalents (Lelley, 1975a,b; Hafiz and Larik, 1984). Thomas and Kaltsikes (1976) concluded that the presence of heterochromatin on certain telomeres of rye chromosomes may inhibit the formation of terminal chiasmata—this results in desynapsis and univalents at MI. In addition, the combination of wheat and rye genomes in the same cytoplasm may cause meiotic irregularities (Larter and Hsam, 1973; Thomas and Kaltsikes, 1976; Hafiz et al., 1982). Thus, cytoplasm or interaction between cytoplasm and nucleus can cause univalents (Larter and Hsam, 1973).

In triticale, another cause of univalency is the DNA content of chromosomes. Kaltsikes (1974) pointed out that each chromosome of rye carries almost 1.5 times as

Table 1
The percentages of cells without univalents at Metaphase I, number of univalents per cell, the percentages of regular Anaphase I cells, and laggard number per cell

Genotypes	Metaphase I		Anaphase I	
	Cells without univalent (%)	Univalent number per cell	Regular cells (%)	Laggard number per cell
High yield				
Nutria 7272	81.4 a*	0.39 a	85.5 a	0.30 a
Iguana 4–2	73.7 cd	0.58 d	82.8 ab	0.41 b
Bgl	77.6 b	0.48 bc	82.2 b	0.42 b
Merino	79.6 ab	0.44 b	80.2 bcd	0.39 b
Hare 2	76.9 bc	0.51 c	78.8 cd	0.50 c
Low yield				
Trasca 87	78.6 ab	0.47 bc	85.8 a	0.30 a
Hare 263	73.3 cd	0.60 d	81.3 bc	0.43 b
Hare 7265	76.5 bc	0.48 bc	77.7 d	0.55 d
Alamos 83	70.1 d	0.72 e	77.6 d	0.63 e
Hare 212	62.7 e	0.90 f	69.4 e	0.86 f
Mean	**75.0**	**0.56**	**80.1**	**0.48**

*Means within columns sharing the same letter do not differ significantly.

much DNA and associated proteins as a wheat chromosome. He postulated that this high amount of DNA in the rye genome requires a much longer time for its meiotic cycle than the tetraploid wheat genome. When wheat and rye are combined in hexaploid triticale, the meiotic cycle of the hybrid is much shorter than rye and this leads to univalent formation.

Anaphase I (AI)

When regular AI cells from high- and low-yielding genotypes were compared, regular AI cells were higher in high-yielding genotypes than low-yielding genotypes. Among anaphase I cells, 80.1% showed regular chromosome segregation (Table 1). The percentage of cells with balanced chromosome segregation (21:21) was highest in Trasca 87 (85.8%) and Nutria 7272 (85.5%), whereas it was lowest in Hare 212 (69.4%).

Mean number of laggards per cell was 0.48. Mean number of laggards per cell was the lowest (0.30) in Nutria 7272 and Trasca 87, whereas it was the highest (0.86) in Hare 212. The cells with two laggards were the most common, followed by the cells with 4, 1, 3, 5, 6 and 7 laggards, respectively.

In this research, the mean number of laggards per cell (0.48) was lower than the mean number of univalents per cell (0.56). This indicated that some univalents segregated to poles without lagging (Hafiz and Larik, 1984; Wang et al., 1989). However, some univalents in MI can cause laggards in AI (Evans and Rahman, 1990). Boyd et al. (1970) noted that the mean frequency of MI open bivalents, univalents, and lagging chromosomes at AI would appear to arise from asynchronous disjunction of chromosomes between MI and AI, resulting in meiotic disturbances and a high frequency of abnormal microspores. The previous studies showed that abnormalities in meiotic chromosome behavior may be generally anticipated in interspecific hybrids and amphidiploids derived from them. The consequences of such abnormalities may be reflected in floret infertility, embryo abortion, incompletely developed seeds, and aneuploidy. The bridges observed in this study may originate from paracentric inversions (Sybenga, 1992).

Anaphase II (AII)

As it is seen in Table 2, 77.1% of all AII cells had regular segregation. The Nutria 7272 had the highest percentage of regular AII cells (83.8%), whereas the Hare 212 had the lowest percentage of regular AII cells (69.4%). There were no laggard(s) in 77.1% of examined cells, but no cells with bridge. With regular AII cells, high-yielding genotypes were higher than low-yielding genotypes.

The mean number of laggards per AII cell for all genotypes was 0.64. The lowest number of laggards per cell was seen in Bgl (0.46), the highest in Hare 212 (0.92). Laggard number per cell varied from 0 to 9 and the most common type was the cell with 2 laggards followed by cells with 1 and 3 laggards.

Table 2
The percentages of regular Anaphase II cells, number of laggard per cell, the percentages of tetrads without micronuclei, the number of micronuclei per tetrad (M/Q), and pollen viability

Genotypes	Anaphase II		Tetrads		Pollen viability (%)
	Regular cells (%)	Laggard number per cell	Tetrads without micronuclei (%)	M/Q	
High yield					
Nutria 7272	83.8 a*	0.47 a	82.8 a	0.52 a	79.8 ab
Bgl	81.1 ab	0.46 a	81.4 a	0.59 b	74.6 bcd
Merino	80.0 b	0.57 b	75.5 bc	0.70 d	74.7 bcd
Iguana 4-2	75.8 c	0.60 bc	74.3 c	0.70 d	80.5 a
Hare 2	71.9 d	0.66 c	66.8 e	0.93 f	78.4 abc
Low yield					
Trasca 87	80.8 ab	0.62 bc	78.0 b	0.66 cd	79.6 ab
Hare 7265	79.6 b	0.61 bc	76.7 bc	0.62 bc	73.8 cd
Hare 263	76.2 c	0.81 d	71.4 d	0.82 e	71.8 d
Alamos 83	72.0 d	0.64 bc	68.4 e	0.98 g	70.0 d
Hare 212	69.4 d	0.92 ef	60.2 f	1.20 h	62.0 e
Mean	**77.1**	**0.64**	**73.5**	**0.77**	**74.5**

*Means within columns sharing the same letter do not differ significantly.

Tetrads

In tetrads, the number of micronuclei was investigated (Table 2). Tetrads at 73.5% had no micronuclei. The percentage of tetrads without micronuclei was highest in Nutria 7272 (82.8%), lowest in Hare 212 (60.0%). On the other hand, the percentage of tetrads without micronuclei in high-yielding genotypes was higher than in low-yielding genotypes.

In Nutria 7272, with the lowest univalent number at MI and laggard number at AI, the micronuclei number per tetrad was also lowest (0.52). The highest number of micronuclei per tetrad was determined in Hare 212 (1.20). Tetrads with 2 and 4 micronuclei were more common than others including 1, 3, 6, and 5 micronuclei, respectively.

The micronuclei in tetrads may originate from fragments or laggards (Pandita and Khoshoo, 1984; Chaubey and Khanna, 1988). It was assumed that tetrads without micronuclei arose from regular meiosis, and there was a negative relationship between micronuclei in tetrads and meiotic stability (Sağsöz et al., 2002). To study meiotic stability, research in tetrads is easier than at other stages of meiosis. This character, therefore, may be used as a selection criterion, especially when a large population is under consideration.

Pollen viability

Pollen viability was examined in the plants used for meiotic studies. In high-yielding genotypes, the percentage of pollen viability was usually higher than in low-yielding genotypes. The percentage of pollen viability was 74.5% (Table 2). The line Iguana 4–2 showed highest pollen viability of 80.5% whereas Hare 212 (62.0%) had the lowest.

Unviable pollens originate from abnormalities in meiosis (Chaubey and Khanna, 1988), cannot fertilize eggs, and cause seed set and grain yield to decrease. Pollen viability may be used as a selection criterion for high seed set (Hwang and Lee, 1992).

The relationships between some cytological characteristics and grain yields

There are negative, significant correlations between grain yield and univalents at MI (r = –0.697), laggards at AI (r = –0.671), AII (r = –0.729), and micronuclei per tetrad (r = –0.648). The correlation between grain yield and chromatin bridge at AI was negative, but not significant. On the other hand, pollen viability affected positively grain yield (r = 0.780) (Table 3).

Our results show that meiotic stability and pollen viability in triticale affect grain yield. Such negative correlations between seed set and meiotic irregularities (univalents, laggards, and micronuclei), have been reported; while meiotic irregularities increased, seed set decreased (Hafiz and Larik, 1984; Melz and Schlegel, 1987; Li et al., 1994).

Table 3
The relationships between some cytological characteristics and grain yields

Correlations	r
Grain yield and	
Number of univalents per MI	–0.697*
Number of laggard per AI	–0.671*
Frequency of AI with bridge	–0.569
Number of of laggard per AII	–0.729*
Micronuclei per tetrad	–0.648*
Pollen viability	0.780**

* and ** indicate significance at 5% and 1% level, respectively.

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