

INTERSPECIFIC HYBRIDIZATION BETWEEN BREAD WHEAT AND AEGILOPS TRIUNCIALIS L.

Laman Namazova^{*}

Genetic Resources Institute of ANAS, Baku, Azerbaijan

Abstract. The possibility for obtaining of interspecific hybrids between bread wheat line (*Triticum aestivum*, 2n = 6x = 42, AABBDD genomes) and different accessions of *Aegilops triuncialis* (2n = 4x = 28, UUCC genome) during two different seasons has been studied. A low crossability rate of 10 % average for all genotypes over two years was achieved. All received F₁ hybrid plants were identical, they exhibited good tillering ability and manifested traits from both parents. In spite of the observed partially fertility in F₁ hybrids between bread wheat and *Aegilops triuncialis* no germination of the hybrid seeds was ascertained. BC₁ seeds were not obtained from F₁ hybrids with bread wheat. Meiotic abnormalities including trivalents were observed suggesting production of unreduced gametes in this cross. During the meiosis of the hybrids, frequencies of chiasmata variated between 1.91and 3.60 per cell.

Keywords: Aegilops triuncialis L., F_1 hybrids, fertility, meiosis.

**Corresponding Author:* Laman Namazova, Genetic Resources Institute of ANAS, Azadlig avenue 155, Baku, Azerbaijan, Tel.: 0558846300, e-mail: <u>leman.namazova.92@mail.ru</u>

Received: 16 October 2019; Accepted: 30 November 2019; Published: 30 December 2019.

1. Introduction

Species belonging to the genus Aegilops L. are an important source of genetic material for expanding genetic variability of cultivated bread wheat, Triticum aestivum L.Thell. (2n = 6x = 42, AABBDD) (Brink & Cooper, 1940). The genus Aegilops comprises 11 diploid and 12 allopolyploid species (Gill & Waines, 1978) with different types of nuclear and cytoplasmic genomes (Grun, 1976). Ae.triuncialis is included in the section Aegilops together with diploid Ae. umbellulata and several polyploid species sharing the U-genome. Ae.triuncialis is subdivided into two subspecies, : ssp. triuncialis and ssp. persica, which carry the same type of nuclear genome, but different cytoplasmic genomes. Ae.triuncialis ssp. persica was originated from hybridization of Ae. umbellulata as female parent with Ae. markgrafii Hammer (syn. Ae. caudata L.), whereas ssp. triuncialis arose from a reciprocal cross (Mann, 1979; Faris et al., 2008). Many accessions of Ae.triuncialis are tolerant to biotic and abiotic stresses. It has been exploited for a wide range of traits including resistance to pests and diseases (Liu et al., 2011; Fetch & Zegeye, 2009; Endo & Tsunewaki, 1975; Endo, 1978; Endo & Gill, 1996; Romero et al., 1998; Dhaliwal et al., 1991; El Bouhssini et al., 1998; Ghazvini, 2012; Makkouk, 1994) and may harbor many other, yet unidentified traits for wheat improvement.

Since the middle of the 20th century, many useful traits have been transferred from alien and progenitor species to wheat (Sears, 1956; Knott & Dvorak, 1976; Kerber 1987).

Ae. triuncialis is an allotetraploid species whose UC genomes are homoeologous to those of *Triticum aestivum* (AABBDD). It is known as the ability conferred by a gene(s) on the C genome, to suppress the Ph diploidization mechanism of *T. aestivum* and *Triticum turgidum*, which normally prevents homoeologous pairing and recombination in polyploidy wheats and their hybrids (Sears, 1976). Previous work has shown pairing among chromosomes of the U and C genomes of *Ae. triuncialis* A-1 and those of *T. aestivum* H-10-15. A cereal cyst nematode (CCN) resistance gene (*Cre7*) was transferred from *Ae. triuncialis* to the TR-353 wheat line (Romero *et al.*, 1998).

The objectives of this paper are to report the results obtained from interspecific hybridization between 171 and 172ACS bread wheat lines and wild species *Aegilops triuncialis*, and as well the characterization of the produced hybrids.

2. Material and methods

The plant materials used in this study were common wheat lines 171ACS and 172ACS ({Aegilotriticale [(T. durumDesf. × *Ae. tauschii*Coss.) × *Secalecereale* L. ssp. segetaleZhuk.]× *T. aestivum* L. 'Chinese Spring'}) and 7 accessions of *Ae.triuncialis* (Girdmanchai, Ismailli, Babek, Nakhchivan, Shamakhı, Shaki and Bolgarıstan) from a collection of Molecular cytogenetics department of AGRI.

In order to obtained intergeneric hybrids between wheat and aegilops used standart methods. Recipient plants have been emasculated according to commonly accepted rules and pollinated by donor plants' pollen (Gorin *et al.*, 1968). The number of seeds set on bread wheat spikes was counted 16-20 days after pollination. Hybrid seeds were cultivated in Petri dishes to check germination ability in the autumn. After the germination ability of the seeds were identified, seedlings were transplanted into the experimental field and phenological observations were made on hybrid plants during vegetation period.

For cytological investigation of meiosis spikes from hybrid plants with anthers containing pollen mother cells were fixed (Tikhomirova, 1990). PMC sprepared by means of the standard Carnoy fixative and acetocarmine squash method.

The study of meiosis in pollen mother cells carried out as follows: anthers have taked from the spikelet flower and placed in special containers which have acetocarmin solution containing in it for coloring. This material was stored in the fridge and was heated several times during the day. Then, temporary slides were made from colored anthers and were studied on a light microscope Motic (China). The results obtained during the study of the meiosis process have been mathematically and statistically developed (Dospekhov, 1979; Lakin, 1990).

3. Results

The results of our crosses are summarized in Table 1 and 2. Crossing success between 171ACS and *Ae. triuncialis* was varied from 2.17 to 9.46 depend on accession of wild parent.

As expected, the F_1 plants were uniform in morphology and demonstrated traits that were obviously inherited from *Ae. triuncialis*, such as spike length, purple color of internodes, and awn length. Other traits, such as number of spikelets per spike and length or width of flag leaf, were similar to those of 171ACS. Compared with the

parents, the F_1 plants had more tillers and showed greater resistance to leaf rust, stem rust, and powdery mildew than did common wheat.

N⁰		Seed setting,	Fertility,	Height,
	Cross combinations	%	%	(cm)
1.	171ACS ×Ae. triuncialis (Girdmanchai, Ismailli)	2.70	steril	66
2.	172ACS ×Ae. triuncialis (Girdmanchai, Ismailli)	9.46	steril	82
3.	171ACS ×Ae. triuncialis (Babek, Nakhchivan)	2.17	steril	91
4.	171ACS ×Ae. triuncialis (Girdmanchai, Ismailli)	8.33	0.35	78
5.	F_2 171ACS × <i>Ae. triuncialis</i>		9.18	55

Table 2. Study of the meiosis process in F_1 hybrids between common wheat lines and
<i>Ae.triuncialis</i>

Hybrid combinations	ATH	Bivalen ts	Ring bivalents	Rod bivalents	Univale nts	Trivale nts	Chiazma frequency	2n
171ACS ×Ae. Triuncialis (Girdmanchai, Ismailli)	108	2.09±0. 27	0.18±0.12	1.92±2.82	30.62±0. 70	0.06±0.09	2.46±0.53	35
172ACS ×Ae. triuncialis(Gird manchai, Ismailli)	80	3.30±0. 26	-	3.3±0.26	28.10±0. 71	0.10±0.1 4	3.60±0.53	35
171ACS × <i>Ae.</i> <i>triuncialis</i> (Baba k, Nakhchivan)	96	1.64±0. 30	-	1.64±2.72	31.64±0. 71	0.03±0.1 4	1.91±0.34	35
41/18-172ACS ×Ae. triuncialis(Gird manchai, Ismailli)	134	3.20±0. 33	0.15±0.08	3.05±2.44	28.57±0. 57	0.01±0.0 8	3.36±0.29	35

The seed setting between 171ACS and 172ACS lines and 2 populations of *Ae. triuncialis* (Girdmanchai accessions) was 2.70 and 9.46%, respectively. Single hybrid seed obtained from each combination. Although, hybrid plants completed their vegetation period, they were completely sterile. The height of plants was 66 and 82 cm, respectively. A study of the meiosis process from first combination revealed that the number of ring and rod bivalves in that pentaploid F_1 hybrid (2n=5x=35) was 0.18 and 1.92 for each PMCs, respectively, the amount of trivalents 0.06 and 30.62 univalents Accordingly, the choromosome frequency was also very low and averaged 2.46, which means that the chromosome conjugation rate is very low. Regarding to the second combination, the number of rod bivalves in that hybrid was 3.30 for each PMCs, the amount of trivalents 0.10 and 28.10 univalents. The choromosome frequency was also very low, approximatedly 3.60.

Only one hybrid seed obtained from combination 171ACS with *Ae. triuncialis* (Babak, Nakhchivan accession) and fertility was 2.17%. This seed germinated and gave F_1 hybrid plant. 91 cm in height, this plant was sterile. During the study of meiosis process of F_1 plants, the number of rod bivalents for each PMCs was 1.64, the number of univalent was 31.64, the amount of trivalent was 0.03, and chiazma frequency was approximately 1.91.

Although, we obtained 1-3 hybrid seeds from combinations between bread wheat line 171ACS and 3 different accessions of *Ae. triuncialis* (Shamakhi, Sheki, and Bulgaria), they could not germinated. Additionally, back-ross hybridization of F_1 hybrids with 171 and 172 ACS lines, cv. Zmitra, N500 line, cv. Grekum 75/50 was failed. It must be cautioned that the seed setting does not always mean success in obtaining F_1 plants.

Regardless of the degree of self-fertility, all of the above pentaploids, at least partially fertile hybrid obtained, when bread wheat crossed with Girdmanchai accession of *Ae. triuncialis* as female parent. Thus, 3 of the 5 hybrid seeds gave F_1 hybrid plants. The height of this plants was about 78 cm and fertility was 0.35%. **During the study of meiosis process of hybrid plants, the number of ring and rod bivalents was 0.15 and 3.05, respectively, the number of univalent was 28.57, the amount of trivalent was 0.01, and chiazma frequency was approximately 3.36. The seedlings from the shriveled seeds were extremely weak and resembled irradiated seedlings in their appearance.Two of the four seeds from F_1 hybrids germinated and gave F_2 hybrids. The height of the plant was 55 cm and fertility was 9.18%.**

4. Discussion

In this study, we successfully produced hybrids between common wheat and *Ae*. *triuncialis*. Observation of morphology revealed that the F_1 hybrids possess characteristics from both parents. Some agronomic traits in the F_1 plants, like spike length, and awn length were similar to those of *Ae*. *triuncialis*, whereas the number of spikelets per spike and the length or width of the flag leaf were similar to those of 171ACS. The size and shape of the seeds of the F_1 differed from those of the male parent *Ae*. *triuncialis* and resembled those of common wheat. Secondly, meiosis in the F_1 hybrids showed that they possessed 2n = 35 chromosomes. Chromosome pairing of the hybrids showed much lower frequency of meiotic pairing, with an average of 2.56 bivalents per cell. Owing to their unbalanced chromosome number and the lack of chromosome pairing, the F_1 plants had poor fertility or sterility with a low seed set rate.

The three most important barriers to wide hybridization are incompatibility (incongruity) between parent species, inviability of the F_1 hybrid and sterility of the F_1 hybrid or its progeny. Cross-incompatibility preventing fertilization arises when pollen grain does not germinate or pollen tube does not reach ovary or male gamete does not fuse with the female gamete. The inviability or weakness of the F_1 hybrid can be due to disharmonies between genomes of parental species, between genome(s) of one species and cytoplasm of other, or between genotypes of F_1 zygote and the genotype of endosperm or maternal tissue (Brink & Cooper, 1940; Gill & Waines, 1978). Deleterious nuclear-cytoplasm interactions have been discussed by Grun (1976) and Maan (1979). When in the F_1 hybrid chromosomes do not pair, gametes receive different number of chromosomes leading, in general, to sterility. Attempts can be made to overcome incompatibility through the choice of parental population, manipulation of parents and emasculation and pollination procedures; to overcome inviability through the application of flower and fruit setting hormones and embryo culture; and to overcome sterility by chromosome doubling or backcrossing of the F_1 hybrid.

The successful use of wheat rust resistance genes previously derived from Aegilops species has encouraged more investigation on these species to discover and exploit new sources of resistance against Ug99 (Stem Rust Pathotype). Several novel

sources of resistance to race Ug99 from Aegilops species of wheat have been recently identified (Faris *et al.*, 2008; Liu *et al.*, 2011). Line Tr129, which was developed by Aung and Kerber (1994) and contains at least one *Aegilops triuncialis* translocation, was found to be resistant to Ug99 in previous work (Fetch & Zegeye, 2009).

Tsujimoto (1984) reported that, chromosome 3C of Aegilops triuncialis, in the monosomic state, causes semisterility in common wheat cultivars (Triticum aestivum). The progeny of plants that carry a single chromosome 3C exhibit chromosome aberrations, and possibly mutations, at high frequencies. Thus, the gametocidal gene on chromosome 3C causes a syndrome similar to hybrid dysgenesis in common wheat. Several alien chromosomes are known to cause semisterility in common wheat when in the monosomic addition state; e.g., chromosome 3C of Aegilops triuncialis (Endo & Tsunewaki, 1975; Endo, 1978). Ae.caudata (Endo & Katayama, 1978), and Ae. cylindrica (Endo, 1978), and chromosome 4s' of Ae. longissima and Ae. sharonensis (Miller et al., 1982). To illustrate, the monosomic addition plants of the common wheat cultivar Triticum aestivum cv. Chinese Spring carrying chromosome 3C of Ae. triuncialis were semisterile because the gametes lacking this chromosome do not function (Endo & Tsunewaki 1975). Additionally, Endo and Gill (1996) have shown that when a particular chromosome from Ae. cylindrica or Ae. triuncialis was present in T. aestivum cv. Chinese Spring in a monosomic condition, chromosomal breakages occurred in the gametes without the alien chromosome, generating various aberrations including deletions. This would explain why we found total sterility in the plants from a cross T. aestivum \times Ae. triuncialis that had been done simultaneously to that of Ae. triuncialis.

The cereal cyst nematode (*Heterodera avenae*) is an important root parasite of common wheat. Romero et al. (1998) reported a high level of resistance was transferred to wheat from *Aegilops triuncialis* (TR lines) using the cross [(*T. turgidum* \times *Ae. triuncialis*) \times *T. aestivum*]. The crosses of line TR-353 with several wheat's cultivars and breeding lines showed that it is possible to produce a sufficient number of viable and fertile progeny for efficient gene transfer. The single resistance factor derived from *Ae. triuncialis*, which it is tentatively designated as CreAet, may represent a new source of genetic resistance to *H. avenae* available to wheat breeders.

Panayotov (1980) reported the most specific in this group was the *Ae. triuncialis* cytoplasm, of which nucleus substitution lines had better growth till heading than control lines. Male sterility was very well expressed with 'Siete Cerros 66', stigmas did not have filaments and anthers were flat with a few or no viable pollen. Pistillody occurred mainly in the top of the 60 spike. Male sterility was not complete in 'Penjamo 62', showing 0.0-29.9% selfed seed fertility. Mean pollen fertility was 26.2% for this line. Apparently 'Penjamo 62' 20 possesses some fertility restoring gens for *Ae. triuncialis* cytoplasm.

Aghaee et al. (2001) presented data that the *Ae. triuncialis* chromosome $5U^t$ has a gene that confers resistance to leaf rust at the seedling stage. They recovered only one recombinant chromosome where the distal part of the short arm was derived from 5AS of wheat and the remaining part of the short arm and the complete long arm was derived from chromosome $5U^t$. Because this recombinant chromosome is mostly derived from $5U^t$ and may contain many agronomically undesirable genes, further chromosome manipulations are needed before this germplasm can be exploited in wheat improvement.

Previously, rust resistance of *Ae. triuncialis* was transferred to wheat using the induced homeologous pairing effect of the PhI gene (Aghaee-Sarbarzeh *et al.*, 2002). Genomic in situ hybridization (GISH) and simple sequence repeat (SSR) marker analysis identified only one leaf rust resistant wheat – *Ae. triuncialis* recombinant, consisting of most of the complete $5U^t$ chromosome with a small terminal segment derived from 5AS (Aghaee-Sarbarzeh *et al.*, 2002). Rust resistance of *Ae. triuncialis* also was transferred to wheat without inducing homeologous pairing between chromosomes of wheat and *Ae. triuncialis* (Harjit-Singh *et al.*, 2000; Aghaee-Sarbarzeh *et al.*, 2001). In one leaf rust resistant line, an introgressed *Ae. triuncialis* segment was identified on chromosome arm 4BS (Aghaee-Sarbarzeh *et al.*, 2001).

Endo and Tsunewaki (1975) informed the cytoplasm substitution lines of three common wheats, i.e., Triticum aestivum cv. Jones Fife (JF), cv. Selkirk (Sk), and T. spelta var. duhamelianum (Spl) with the cytoplasm of Aegilops triuncialis showed partial male fertility (about 25 percent on the average) and remarkable female sterility (about 80 percent), and those with the cytoplasm of synthetic triuncialis produced from Ae. caudata × Ae. umbellulata, showed complete male sterility and very high female sterility (about 90 percent). The female and male sterility was associated with the preferential transmission of a subterminal chromosome (i chromosome) of Ae. triuncialis and synthetic triuncialis through the gametes of both sexes. Selfed progeny of the three cytoplasm substitution lines with Ae. triuncialis cytoplasm had two i chromosomes without exception and restored both male and female fertility to an almost normal level. It is proposed that gametophyte sterility is caused by the inviability of the gametes in plants lacking the i chromosome, regardless of the kind of cytoplasm possessed by the plants. Conversely, the gametes carrying the i chromosomes are functional in the same plants, resulting in the preferential transmission of this chromosome to the offspring.

5. Conclusion

At interspecific hybridization between bread wheat genotypes as female parent and three accession of wild relative *Aegilops triuncialis* was achieved low crossability rate of 5.70 % average for all genotypes. All received F_1 hybrid plants from the cross $171ACS \times Aegilops$ triuncialis were identical, exhibited good tillering ability and manifested traits from both parents. Here presented results are only initial step of the involvement of wild species *Ae. triuncialis* in long process of production of bread wheat breeding lines with introgressed alien genes. Successive progenies are going to be screened at morphological, physiological, cytological and molecular level for hybrid identification and enhancing of genetic variation for biotic - and abiotic stress resistance traits and its incorporation into common wheat.

References

Gorin, A.P., Dunin, M.S., Konovalov, Yu.B. et al. (1968). Workshop on selection and seed production of field crops. Moscow: Kolos, 439 (in Russian).

Dospekhov B.A. (1979). Field Experience Technique. Moscow: Kolos, 416 (in Russian).

Lakin, G.F. (1990). Biometrics. Moscow: Higher School, 352 (in Russian).

Tikhomirova, M.M. (1990) *Genetic analysis*. Textbook allowance. Publishing house of LSU, 280 (in Russian).

- Aghaee-Sarbarzeh, M., Harjit-Singh, & Dhaliwal, H.S. (2001). A microsatellite marker linked to leaf rust resistance transferred from *Aegilops triuncialis* into hexaploid wheat. *Plant Breed.*, 120, 259–261.
- Aghaee-Sarbarzeh, M., Ferrahi, M., Singh, S., Harjit-Singh, Friebe, B., Gill, B.S., & Dhaliwal, H.S. (2002). PhI induced transfer of leaf and stripe rust resistance genes from *Aegilops triuncialis* and *Ae. geniculata* to bread wheat. *Euphytica* 127, 377–382.
- Aung, T. & Kerber, E.R. (1994). Incorporation of leaf rust resistance from wild tetraploid into cultivated hexaploid wheat. *Ann. Wheat Newslet.*, 40, 83-84.
- Brink, R.A. & Cooper, D.C. (1940). Double fertilization and development of the seed in angiosperms. *Bot.Gaz.*, 102, 1-25.
- Dhaliwal, H.S., Harjit-Singh, S. Gupta, P.S. Bagga, & K.S. Gill.(1991). Evaluation of Aegilops and wild Triticum species for resistance to leaf rust (*Pucciniareconditaf.sp. tritici*) of wheat. *Int. J. Trop. Agric.*, 9, 118–121.
- Endo, T.R. & Tsunewaki, K. (1975). Sterility of Common Wheat With Aegilops triuncialis Cytoplasm. The Journal of Heredity, 66, 13-18.
- Endo, T.R. (1978). On the Aegilops chromosomes having gametocidal action on common wheat. *Proc. 5th Int. Wheat Genet. Symp.*, 306-314.
- Endo, T.R., Gill, B.S. (1996). The deletion stocks of common wheat. J. Hered., 87, 295-307.
- Endo, T.R., Katayama, Y. (1978). Finding of a selectively retained chromosome of *Aegilops* caudata L. in common wheat. *Wheat Inf. Serv.*, 47-48, 32-35.
- El Bouhssini, M., Benlhabib, O., Nachit, M.M., Houari, A., Bentika, A., Nsarellah, N., &Lhaloui, S. (1998). Identification in Aegilops species of resistant sources to *Hessian fly*. *Genet. Res. Crop. Evol.*, 45, 343–345.
- Faris, J.D., Xu, S.S., Cai, X., Friesen, T. L. & Jin, Y. (2008). Molecular and cytogenetic characterization of a durum wheat-Aegilops speltoides chromosome translocation conferring resistance to stem rust. Chromosome Res., 16.
- Fetch, T. & Zegeye, T. (2009). Inheritance of resistance to Ug99 in wheat line Tr129 with an introgression of Aegilops triuncialis chromatin. Proceedings of 12th International Cereal Rusts and Powdery Mildews Conference. Antalya, Turkey, 33.
- Ghazvini, H., Hiebert, C.W., Zegeye, T., & Fetch T. (2012). Inheritance of stem rust resistance derived from *Aegilops triuncialis* in wheat line Tr129. *Can. J. Plant Sci.*, 92, 1037–1041.
- Gill, B.S., Waines, J.G. (1978). Paternal regulation of seed development in wheat hybrids. *Theor. Appl. Genet.*, 51, 265-270.
- Grun, P., (1976). Cytoplasmic genetics and evolution. Columbia Univ. Press, NY.
- Harjit-Singh, & Dhaliwal, H.S. (2000). Intraspecific genetic diversity for resistance to wheat rusts in wild Triticum and Aegilops species. *Wheat Inf. Serv.*, 90, 21–30.
- Kerber, E.R. (1987). Resistance to leaf rust in hexaploid wheat, Lr32, a third gene derived from *Triticum tauschii. Crop Sci.*, 27, 204-206.
- Knott, D.R., Dvorak, J. (1976). Alien germplasm as a source of resistance to disease. *Annu. Rev. Phytopathol.*, 14, 211-235.
- Liu, W. X., Jin, Y., Rouse, M., Friebe, B., Gill, B. & Pumphrey, M.O. (2011). Development and characterization of wheat- *Ae.searsii* Robertsonian translocations and a recombinant chromosome conferring resistance to stem rust. *Theor. Appl. Genet.*, 122.
- Makkouk, K.M., Comeau, A., & Ghulam, W. (1994). Resistance to barley yellow dwarf luteovirus in *Aegilops* species. *Can. J. Plant.Sci.*, 74, 631–634.
- Mann, S.S., (1979). Specificity of nucleo-cytoplasmic interactions in *Triticum* and *Aegilops* (a review). *Wheat Info. Serv.*, 50, 71-79.
- Martin-Sanchez, J.A., Gomez-Colmenarejo, M., Del Moral, J., (2003). A new *Hessian fly* resistance gene (H30) transferred from the wild grass *Aegilops triuncialis* to hexaploid wheat. *Theor. Appl. Genet.*, 106, 1248–1255.
- Miller, T.E., Hutchinson, J. & Chapman, V. (1982).Investigation of a preferentially transmitted *Aegilops sharonensis* chromosome in wheat. *Theor. Appl. Genet.*, 61, 27-33.

- Panayotov, I. (1980). New Cytoplasmic Male Sterility Sources in Common Wheat: Their Genetical and Breeding Considerations. *Theor. AppL Genet.*, 56, 153-160.
- Romero, M.D., Montes, M.J., Lopez-Branda, E., Duce, A., Martin-Sanchez, J. A., Andres, M. F., & Delibes, A. (1998). A cereal cyst nematode (*Heteroderaavenae* Woll.) resistance gene transferred from *Aegilops triuncialis* to hexaploid wheat. *Theor.App. Genet.*, 96, 1135-1140.
- Sears, E.R. (1956). The transfer of leaf rust resistance from *Aegilops umbellulata* to wheat. Brookhaven Symp. Biol. 9, 122.
- Tsujimoto, H., Tsunewaki, K. (1984). Gametocidal genes in wheat and its relatives. I. Genetic analyses in common wheat of a gametocidalgene derived from *Aegilops spetoides. Can. J. Genet.Cytol.*, 26, 78-84.