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PRODUCTION OF HYBRIDS BETWEEN *T. AESTIVUM* L. AND *AE. VENTRICOSA* TAUSCH

Research article

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Abstract

The purpose of the current study was to estimate the crossing ability and chromosome pairing behavior of intergeneric F₁ hybrids between *Ae. ventricosa* (D^vD^vN^vN^v) and *Triticum aestivum* (AABBDD) crosses. During the production of *T. aestivum* – *Ae ventricosa* hybrids, it was observed that F₁ hybrids' morphological traits were intermediate between parents, the plants grew vigorously but were completely sterile. Seed set ranging from 2.17 to 8.00 %. Hybrids (2n = 5x = 35) produced without embryo rescue methods. The present study revealed that, the average frequency of chromosome associations at metaphase I was averaged 25.39 univalents and 4.78 bivalents. All received F₁ hybrid plants were identical, they exhibited good tillering ability and manifested traits from both parents.

Keywords: *T. aestivum*, *Ae. ventricosa*, intergeneric hybridization, meiosis.

МЕЖРОДОВАЯ ГИБРИДИЗАЦИЯ *T. AESTIVUM* L. И *AE. VENTRICOSA* TAUSCH

Научная статья

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Аннотация

Во время гибридизации между *T. aestivum* и *Ae ventricosa* было обнаружено, что морфологические признаки гибридов F₁ были промежуточными между родительскими формами, что растения сильно росли, но были полностью стерильными. Завязываемости семян в гибридных комбинациях варьировалась от 2,17 до 8,00%. Гибриды (2n = 5x = 35), полученные без методов спасения эмбрионов. В результате мейотических анализов выявлено, что среднее число унивалентов было 25,39, а бивалентов было 4,78. Все полученные гибридные растения F₁ имели хорошую разветвленность и имели признаки обоих родителей.

Ключевые слова: *T. aestivum*, *Ae. ventricosa*, межродовая гибридизация, мейоз.

Introduction

The tetraploid species *Aegilops ventricosa* Tausch (syn. *Triticum ventricosum*) belongs to the tribe Triticeae. It is closely related to wheat (*Triticum aestivum* L.) (2n = 6x = 42, AABBDD) since its genomic composition is D^vD^vN^vN^v, with the D^v and N^v genomes partially homologous to the D genome of *Aegilops tauschii* L. and to the N genome of *Aegilops uniaristata* Vis., respectively [22]. It carries many valuable genes, such as resistance to eyespot [10], cereal cyst nematode [14], [10], Hessian Xy [8], leaf, yellow and stem rust [1], [19], and karnal bunt [21]. Mapping of the rust resistance genes *Yr17*, *Lr37*, and *Sr38* using molecular markers revealed the presence of a small translocation between the *Ae. ventricosa* chromosome 2N^v and chromosome 2AS of bread wheat [1].

Dosba et al. (1978) produced a set of wheat – *Ae. ventricosa* disomic addition lines [6]. The added pair in one of them was easily identified in mitotic preparations because it was subtelocentric [7]. Both its sporophytic and gametophytic compensations for 6D were high, so the initial expectation was that the *Ae. ventricosa* pair would prove to be fully homoeologous with wheat homoeologous group 6. It was thus designated as 6N^v.

Genetic recombination occurs when the parental genomes share enough homology for homoeologous chromosomes to pair and recombine during meiosis. Chromosome pairing in the interspecific hybrid may occur in two forms: (1) autosyndesis, pairing occurs between members of the same parental genome, and (2) allosyndesis, pairing occurs between members of different parental genomes [2]. If chromosomes behave as univalents, do not pair during meiosis, or pair autosyndetically DNA is not exchanged between genomes and introgression through genetic recombination will not be possible. A requisite amount of homology between the different genomes will allow homoeologous chromosomes to recombine during meiosis, resulting in recombinant chromosomes carrying both donor and recurrent parent genetic material. All forms of introgression have been successful in transferring agronomically useful traits from wild relatives into cultivated species [15]. Interspecific hybridization can greatly increase the amount of genetic variation available to plant breeders for improvement.

The aim of this study was to investigate the crossing ability and morphology of wheat – *Ae. ventricosa* hybrids and their meiosis behaviour.

Material and methods

For the material of the study 2 common wheat lines were used – 171 and 172ACS ($\{Aegilotriticale [(T. durum Desf. \times Ae. tauschii Coss.) \times Secale cereale L. ssp. segetale Zhuk.] \times T. aestivum L. 'Chinese Spring', N500 [(T. aestivum var. graecum Suriya \times Ch. Spring) \times cv. Bezostaya-1]$, and 2 *Ae. ventricosa* accessions.

The emasculation and pollination in the field were carried out during the months of April-May in 2019 at the Absheron Research Base of Genetic Resources Institute. No embryo rescue or hormone treatment was applied for the production of F₀ seeds. The spikes were harvested and the number of seed-set per spike was counted about 20 days after pollination. Crossability was expressed as the percentage of the number of seed set to the total number of florets pollinated.

For meiotic studies, young spikes at the appropriate stages were fixed in Carnoy's fixative for 24 h and then stored in 70% ethanol. Pollen mother-cells were stained and squashed in 1% acetocarmine. Meiotic observations were made and documented with Motic (China) microscope.

Results

Table 1 - Crossing ability of *Ae. ventricosa* with *T. aestivum*

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№	Cross combinations	Seed setting, %	Obtained seed, absolute number	Plant, absolute number
1.	171ACS \times <i>Ae. ventricosa</i>	2.17	1	-
2.	171ACS \times <i>Ae. ventricosa</i> TA-1999	6.94	5	3
3.	172ACS \times <i>Ae. ventricosa</i> TA-1999	-	-	-
4.	<i>Ae. ventricosa</i> \times 172ACS	8.00	4	-
5.	172ACS \times <i>Ae. ventricosa</i>	4.35	2	2



Figure 1 - Spike of hybrid 171ACS \times *Ae.ventricosa* (center), bread wheat parent (left) and wild parent (right)

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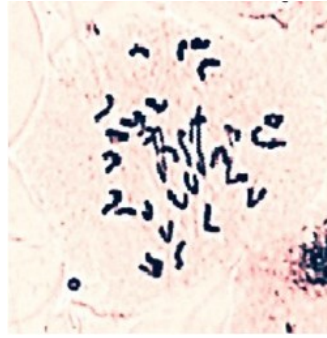


Figure 2 - Meiotic behaviour of F₁ plant
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Seed setting between common wheat line 171ACS and *Ae. ventricosa* was 2.17%, and cross with *Ae. ventricosa* TA-1999 was 6.94%. Obtained single seed from the first combination did not germinate. Regarding the next hybrid, 3 of the 5 seed germinated and gave F₁ plants. The height of the plants was 101 cm and were completely sterile. Thus, it was not possible to get any seed from 1624 spikelet flowers. During the study of meiosis process of F₁ plants, the number of ring and rod bivalents for each PMCs (pollen mother-cells) was 0.54 and 4.37, respectively, the number of univalent was 25.18, and chiasma frequency was approximatedly 5.44 (table 2).

Reciprocal crosses carried out between 172ACS and *Ae. ventricosa*. Seed setting of combination when *Ae. ventricosa* TA-1999 used as a female parent was 8.00%, however obtained 4 seeds did not germinate. Seed setting of 172ACS × *Ae. ventricosa* combination was 4.35%. Obtained 2 seeds germinated and gave hybrid plants that had intermediate morphotype. The height of the plants was about 103 cm and were completely sterile. Thus, it was not possible to get any seed from 1892 spikelet flowers. During the study of meiosis process of hybrid plants, the number of ring and rod bivalents for each PMCs was 1.75 and 2.89, respectively, the number of univalent was 25.60, the amount of trivalents 0.04 and chiasma frequency was approximatedly 6.47.

Backcross hybridizations between F₁ plants and different bread wheat lines (171ACS, N500, cv. *Siete cerros* and cv. *Zmitra*) was failure.

Kihara (1949) reported the three plants of the cross "Courttdt" × *Ae. ventricosa* I had 2n = 34 chromosomes (5B absent) and, of those resulting of the crosses "Chinese Spring" × *Ae. ventricosa* II, one had 34 chromosomes and the other two 35 chromosomes. They grew quite normally and were intermediate to their parents in most morphological characters. The hybrid plants were sterile, and the seed setting was very low [11]. These results were similar with ours.

Table 2 - Study of the meiosis process in F₁ hybrids between common wheat lines and *Ae. ventricosa*

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Hybrid combinations	ATH, absolute number	Bivalents, absolute number	Ring bivalents, absolute number	Rod bivalents, absolute number	Univalents, absolute number	Trivalents, absolute number	Chiasma frequency, absolute number
171ACS × <i>Ae. ventricosa</i>	142	4.91±0.45	0.54±0.11	4.37±1.46	25.18±0.85	-	5.44±6.58
172ACS × <i>Ae. ventricosa</i>	97	4.64±0.28	1.75±0.40	2.89±1.78	25.60±0.88	0.04±0.22	6.47±0.91

Discussion

Obtaining interspecific hybrids offers significant variability. It is the crucial step in any program of genes introgression from wild species. However, its success depends not only on the choice of genitors that suits the objectives of the program, but also on crossing affinity.

Previous works had shown the effectiveness of a method to transfer genes from the wild grass *Ae. ventricosa* to the cultivated wheat *T. aestivum* [4], [13]. The strategy involves a cross of the donor species, *Ae. ventricosa* with *T. turgidum* (AABB), which acts as a bridge, followed by the rescue of the sterile ABD^vM^v hybrid with pollen from the recipient species *T. aestivum* (AABBDD). Plants from this cross were fertile and stable lines with 42 chromosomes were derived from them after repeated selfing.

The few works dedicated to interspecific crosses where species of the genus *Aegilops* are taken as female parent, report the weakness of obtaining such hybrids, which seems, more favourable in field conditions [9]. Many criteria influence the acquisition of fertile hybrids and backcross progenitor for introgression between two genera, including genetic relationships, ploidy level, and hybridization direction [20]. The success of obtaining hybrids depends largely on the parental genotypes involved in the crossing. The differences in hybridization affinity of *Aegilops* species and wheat varieties are highly observed

[9], [16], [20]. In many studies, the common sharing of the D genome between the bread wheat and the wild parent allowed the pairing of homeologous chromosomes and obtaining fertile hybrids [17], [18].

The distribution of biochemical markers and the maximum number of chromosomes from the M^v genome in the selected lines obtained by Olmedo [9]. The latter is deduced from the number of univalents at MI observed in the hybrid and represent an overestimation for two reasons:

I) because it has been repeatedly observed that there is incomplete homology between the D genomes of *T. aestivum* and *Ae. ventricosa*, and

II) because the transfer of a terminal segment of a chromosome from the M^v genome to a wheat chromosome would suffice to drastically reduce its meiotic pairing with the original wheat chromosome.

It can be concluded, from the joint consideration of the biochemical and the cytological data, that the genetic transfer from the M^v genome of *Ae. ventricosa* to hexaploid wheat has taken place not only by chromosome substitution but also by recombination. In other words, lines carrying, at most, one whole M^v genome chromosome also carry two independently inherited M^v genome markers.

E. D. Badayeva informed that hybridization of species that share common genomes can induce an extensive exchange of genetic material between closely related chromosomes. According to meiotic analysis, the D-genome of *Ae. ventricosa* is only slightly modified relative to its diploid progenitor [12]. The D-genome of common wheat is also very similar to *Ae. tauschii*. We might expect that wheat–*Ae. ventricosa* D-genome chromosome pairing could have occurred in the F₁ hybrid plant, giving rise to recombinant D-D^v genome chromosomes. Indeed, a high level of pairing was observed in wheat–*Ae. ventricosa* hybrids [9]. In contrast, the N^v genome is quite distant from the A- and B-genomes, and they rarely pair at meiosis [3]. These results underline the use of complementary markers, such as biochemical, molecular, and cytological markers in the production and characterization of addition and introgression lines, particularly if the parental species share a common genome which interferes with meiotic pairing in hybrids.

Conclusion

At interspecific hybridization between bread wheat genotypes and two accession of wild relative, *Aegilops ventricosa* was achieved with low crossability rate for all genotypes. Here presented results are only the initial step of the involvement of wild species *Ae. ventricosa* 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. All wheat-aegilops hybrids involved backcross hybridization, but it was ineffective.

Конфликт интересов

Не указан.

Рецензия

Все статьи проходят рецензирование. Но рецензент или автор статьи предпочли не публиковать рецензию к этой статье в открытом доступе. Рецензия может быть предоставлена компетентным органам по запросу.

Conflict of Interest

None declared.

Review

All articles are peer-reviewed. But the reviewer or the author of the article chose not to publish a review of this article in the public domain. The review can be provided to the competent authorities upon request.

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