Evidence of Homoeologous Relationship Between Chromosomes of Wheat and of *Aegilops geniculata*

M. Asghar, A. R. Rao and S. Farooq

1Nuclear Institute for Agriculture and Biology (NIAB), P. O. Box 128 Faisalabad, Pakistan
2Department of Botany, University of Agriculture, Faisalabad-38040, Pakistan

Abstract: Intergeneric hybrids were produced between hexaploid wheat variety Chinese Spring and five different accessions of *Aegilops geniculata*; 2n = 4x = 28 C'C'M'M'M' (Aegilops ovata). The objective was to detect differences in chromosomal associations at meiotic metaphase-I in the F₁ hybrids using squash preparations of prefixed immature anthers. Chromosome pairing between all the hybrids ranged between 36 univalents to 3 rod bivalents except for hybrid between Chinese Spring × *Ae. geniculata* accession 361881. This hybrid exhibited chromosome pairing up to 4 rod bivalents, 1 ring bivalent and 2 trivalents with the chiasma frequency of 4.10 per cell. This indicated possibilities of homoeologous chromosome pairing and genetic exchange between chromosomes of wheat and of *Ae. geniculata*. Significance of this pairing and the potential of *Ae. geniculata* for improvement of stress tolerance in wheat would be discussed in detail.

Key words: Intergeneric hybrids, *Triticum aestivum, Aegilops geniculata*, Homoeologous pairing, Chiasma frequency

Introduction

Meiotic chromosome pairing in hexaploid wheat is under strict control of pairing promoter genes located on homoeologous group 3 and 5 (Feldman, 1966) while its suppressers are located on the long arm of chromosome 6B (Sears and Okamoto, 1968) and short arm of chromosome 3D (Sears, 1963). Restriction of meiotic pairing to fully homologous chromosomes is mainly due to the effect of a strong homoeologous pairing suppressor gene located on the long arm of chromosome 6B (6B L) later designated as Ph (pairing homoeologous) by Wall et al. (1971) and presently Ph-1 by McIntosh (1979). When this gene is present even hemizygotically, only homologous chromosome can pair. However, meiotic studies made in different intergeneric hybrids (Charpentier et al., 1988 and Farooq et al., 1990a) indicated that if the alien species possesses the genes that are epistatic to wheat pairing control system, homoeologous pairing is expected to occur even in the presence of Ph genes. In one of such studies made between hexaploid wheat and different accessions of *Ae. variabilis* (2n = 4x = 28 C'C'S'S') accessions A, B, and E (Farooq et al., 1990b), higher chiasma frequency was exhibited by hybrids involving accession E than that of A and B.

*Ae. geniculata*; 2n = 4x = 28 C'C'M'M'M' (Aegilops ovata) is another tetraploid species which shares one genome (C') with *Ae. variabilis*. It possesses resistance (Warham et al., 1969) against Karnal bunt (*Neovesia indica*) which is one of the serious wheat diseases. Some of its accessions also possess salt tolerance (Farooq et al., 1989; Farooq, 1997; Farooq et al., 1997). Whether accessions of *Ae. geniculata* also possesses pairing promoter genes is not yet known and can be found at meiotic metaphase-I in the F₁ hybrids of wheat with accessions of *Ae. geniculata*. The objective of the present study is therefore, to produce hybrids between different accessions of *Ae. geniculata* with wheat variety Chinese Spring and to study the chromosomal associations between the two species at meiotic metaphase-I.

Materials and Methods

Material used in this study comprised wheat variety Chinese Spring and five different accessions of *Ae. geniculata* L. viz. F, Ciae-65, P1-276978, P1-330487, and P1-361881. Accession F was obtained from Plant Breeding Institute, Cambridge, UK (now Cereal Research Department, John Innes Centre, Norwich, UK), while the other 4 accessions were obtained from USDA, ARS, Beltsville, Maryland, USA. The origin of accession F and Ciae-65 is unknown while that of accessions P1-276978, P1-330487 and P1-361881 is Japan, U.K., and Romania respectively.

Intergeneric hybrids were produced using hand emasculation of wheat variety Chinese Spring used as female parent and pollination with different accessions of *Ae. geniculata*. Post pollination treatment with growth hormones was also applied (Kruse, 1973). Seeds were harvested at maturity after the crossability data was recorded. The hybrid seeds were germinated and root tips were collected from two weeks old seedlings for chromosomes counting. The root tips were pretreated with a solution of 0.05% colchicine and 0.025% 8-hydroxyquinoline for 3 hours and then transferred to 1% acet-o-orcein and stored in refrigerator. Mitotic preparations were made by squashing the root tips in 45% acetic acid. The chromosomes were examined under microscope and were photographed using 6x photo lens of Olympus Vanox-S research microscope. All F₁ plants that showed 35 chromosomes were transferred into 25cm diameter plastic pots filled with soil and were placed in the net house in the month of November. At flowering, immature spikes were fixed in Carnoy's solution. After fixation for two days at room temperature, spikes were transferred to 70% ethanol and were stored in freezer. Anthers with dividing PMCs at metaphase-I were extracted and fixed in alcholic hydrochloric acid carmine (Snow, 1963). Mitotic preparations were made by squashing the anthers in a drop of 45% acetic acid on a glass slide. Cover slips were removed by freezing the material at -70°C in ultra-cold freezer. Photographs were taken in a manner similar to that of mitotic preparations. The present study was carried out at Nuclear Institute for Agriculture & Biology (NIAB), Faisalabad, Pakistan as a part of Plant Molecular Breeding work and completed in two years.

Results

Mitotic analysis of all the hybrids showed 35 chromosomes (Fig. 1a) including 21 of wheat and 14 of *Ae. geniculata* chromosomes. The hybridity was additionally confirmed through plant morphology, which indicated co-dominant phenotype.

Mitotic analysis made at metaphase-I in the hybrids of Chinese Spring with accession 276978 of *Ae. geniculata*
Table 1: Mean meiotic chromosomal associations at metaphase-1 in the F₁ intergeneric hybrids of wheat variety Chinese Spring (C.S.) with Aegilops geniculata

<table>
<thead>
<tr>
<th>Hybrid combination</th>
<th>No. of cells analyzed</th>
<th>Mean meiotic associations</th>
<th>Chiasmata/cell</th>
</tr>
</thead>
<tbody>
<tr>
<td>C.S. x Ae. geniculata acc. 301881</td>
<td>30</td>
<td>28.16 2.24 0.43 0.80</td>
<td>4.10 A</td>
</tr>
<tr>
<td>C.S. x Ae. geniculata acc. 35</td>
<td>30</td>
<td>30.86 0.17 1.40 0.30</td>
<td>2.34 B</td>
</tr>
<tr>
<td>C.S. x Ae. geniculata acc. 330487</td>
<td>30</td>
<td>31.37 0.27 1.30 0.03</td>
<td>2.10 B</td>
</tr>
<tr>
<td>C.S. x Ae. geniculata acc. F</td>
<td>30</td>
<td>33.06 0.10 0.67 0.20</td>
<td>1.17 C</td>
</tr>
<tr>
<td>C.S. x Ae. geniculata acc. 278909</td>
<td>30</td>
<td>33.12 0.06 0.88 -</td>
<td>1.00 C</td>
</tr>
</tbody>
</table>

Figures followed by the same letter are non-significant at 5% level of significance according to DMRT.

Fig. 1: Cytological behaviour in Chinese Spring x Ae. geniculata F₁ hybrids:
- a) Mitotic chromosomes (2n = 6x = 36). Meiotic chromosomal association at metaphase-1 in Chinese Spring with Ae. geniculata
- b) acc. 278909, 36 univalents, 31 univalents, 2 rod bivalents

Fig. 2: Meiotic chromosomal association at metaphase-1 in Chinese Spring with different accessions of Ae. geniculata:
- a) acc. F, 31 univalents, 2 rod bivalents
- bl acc. 66, 29 univalents, 3 rod bivalents
- c) acc. 301881, 27 univalents, 4 rod bivalents
- dl acc. 361881, 27 univalents, 1 rod bivalent, 2 trivalents
rod bivalents (Fig. 2c) to 27 univalents, 1 rod bivalent and 2 trivalents (Fig. 2d) in Chinese Spring x Ae. geniculata acc. 361881. Significant difference in mean chiasma frequencies was also observed among the hybrids of different accessions of Ae. geniculata with Chinese Spring (Table 1). The highest chiasma frequency (4.10) was observed in the hybrid of Chinese Spring and Ae. geniculata accession 361881. There was no significant difference in the chiasma frequencies observed in the hybrids of Chinese Spring and Ae. geniculata accessions 66 and 330487. Similarly, difference in chiasma frequencies observed in the hybrids of Chinese Spring with Ae. geniculata accessions F and 276979 was also not significant.

Discussion
The frequency of different meiotic figures observed in the present study particularly in Chinese Spring x Ae. geniculata accession 361881 is significantly higher than the frequency generally expected in intergeneric hybrids. Normally, such hybrids reveal chromosomal associations of 1-3 rod bivalents except in the hybrids of wheat with alien species where Ph genes are not functional. In the wheat variety Chinese Spring used in the present study, Ph genes were fully active yet, the frequency of rod bivalents is significantly high. This pairing which may or may not be between the chromosomes of wheat and of Ae. geniculata reflects the presence of some additional pairing promoter genes particularly on Ae. geniculata. Such effect have already been observed by Farooq et al. (1999 and 1990b) in different accessions of Ae. variabilis. Since Ae. geniculata shares C1C2C3 genome with Ae. variabilis, it is possible that the gene(s) affecting meiotic pairing in intergeneric hybrid could also be present in Ae. geniculata accession 361881. This was also indicated in the hybrids of wheat cv LU-26 with Ae. geniculata accession 361881 where chiasma frequency as high as 10 per cell was observed (Farooq et al., 1996). The significance of this pairing is that the economically important genes like genes for karnal bunt resistance and for salt tolerance residing in Ae. geniculata could be transferred to cultivated wheat through crossing over between chromosomes of wheat and of Ae. geniculata which is a stable transfer compared to the transfer made through production of addition and substitution lines which are beset with chromosomal and yield instability.

References


