

WELCOME



ASSIGNMENT

ON

Alien Addition Lines & Their Use in Crop Improvement



Submitted To:

Dr. N.K. Sharma

(Prof.) & Director, IABM

Submitted By:

Kapil Bishnoi

M.Sc.(Ag.)Pre., GPB Dept.

Department of Genetics & Plant Breeding

College of Agriculture,

Swami Keshwanand Rajasthan Agricultural University, Bikaner

HYBRIDIZATION

Crossing between two **genetically dissimilar** parents is called hybridization.

□ Wide Hybridization

1. Interspecific hybridization:- Crosses made between distantly related species.

Ex. Nerica, an upland rice

Oryza sativa (Asian rice) X *Oryza glaberrima* (African rice)

Tetraploid species of *Brassica spp.* (**U- triangle given by Nagaharu, 1935.**)

2. Intergeneric hybridization:- Crosses made between distantly related genera

Ex. Triticale, a cross between **wheat** (*Triticum aestivum*) and **rye** (*Secale cereale*)

developed by Rimpu, in Germany(1890) .

Ex. Raphanobrassica, a cross between radish and cabbage.

Why wide hybridization?

- Development of new hybrids for diseases and insect resistance, high quality, wider adaptation, higher yield.
- Development of new variety
- Production of new crop species (e.g. *Triticale*)
- Wider adaptation
- Transfer of cytoplasm

Problems in wide hybridization:

- Poor crossability
- Early embryo abortion
- Hybrid seed inviability
- Hybrid seed lethality
- Hybrid sterility due to low chromosome pairing

Techniques to remove these barriers in distant hybridization :

- Application of growth hormones (IAA) to reduce embryo sac abortion
- Under controlled condition (Tissue culture technique)
- Somatic hybridization
- Restoration of seed fertility by doubling the chromosome of sterile F1 hybrids (colchicine treatment)
- Utilization of bridge crosses where direct crosses are not possible.
- **Alien addition lines**
- **Alien substitution lines**

- Transfer of economically important traits from wild species to the cultivar is achieved by alien gene transfer, which is possible:
 - **At genome level:** Production of amphiploids
(eg. Triticale and raphanobrassica)
 - **At chromosome level:** Production of alien addition and substitution line.
 - **At the level of chromosome segment:** Production of translocation lines.

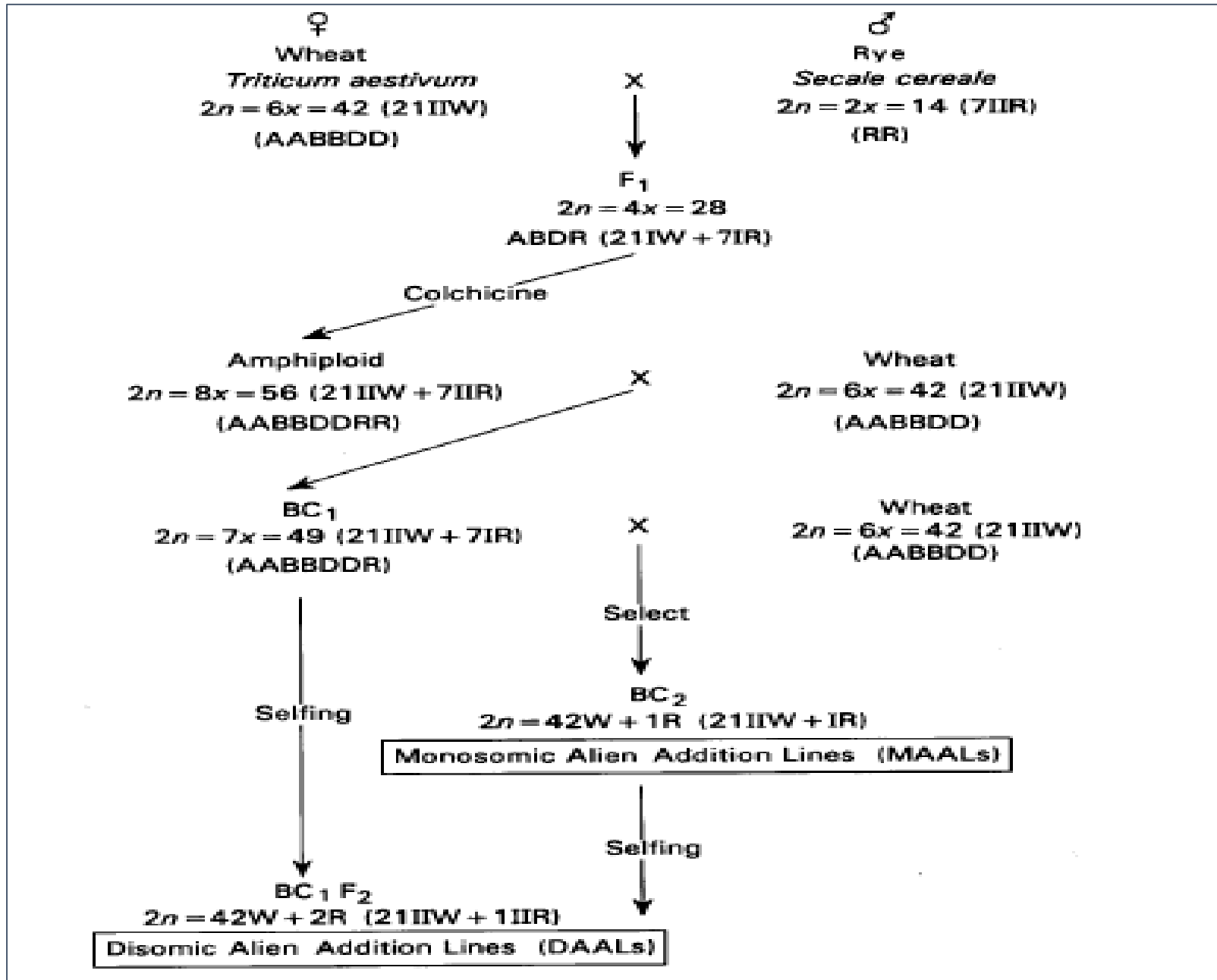
ALIEN ADDITION LINES

- Term given by Clausen (1945).
- These lines carries one chromosome pair from a different species in addition to the normal somatic chromosome complement of the parent species (**$2n+2$ =Alien addition lines**)
- When only one chromosome from another species is present, it is known as **Monosomic Alien Addition Lines (MAALs)**. ($2n+1$ = Monosomic alien addition line)
- Selfing of MAALs produce **Disomic Alien Addition Lines(DAALs)**.
- The main purpose - **transfer of disease resistance from related wild species to cultivated spices.**
- e.g. transfer of mosaic resistance from *Nicotiana glutinosa* to *N. tabacum*.
- MAALs have been produced in wheat, rice, oats, tobacco, cotton, soybean and several other species.
- Procedure described by **O'Mara (1940)**

General Procedure:-

- **Production of inter-specific or inter-generic F1 hybrid**
- **Induction of an amphiploid**
- **Generation of BC1**
- **Isolation of MAALs in BC2 & BC3**
- **Selection of DAALs after selfing of the MAALs.**

Procedure for obtaining MAALs & DAALs of wheat-rye



- Hexaploid wheat and diploid rye crosses result in F1 polyhaploid sterile plants ($2n = 4x = 28$, ABDR)
- The octoploid ($2n = 8x = 56$, crosses AABBDDRR) triticales is synthesized by doubling the chromosomes with colchicine.
- Octoploid triticales is fertile and is backcrossed to wheat giving heptaploid plants with $2n = 7x = 49$, AABBDDR.
- Heptaploid plants exhibit $21IIW + 7IR$.
- The seven rye univalents are randomly excluded or included in the gametes.
- MAALs ($2n = 42W + 1R - 7R$) are isolated either after selfing the heptaploid or the heptaploid is again crossed by wheat.
- DAALs are isolated in the selfed population of MAALs.

IDENTIFICATION OF ALIEN ADDITION LINES

Alien addition lines can be identified by

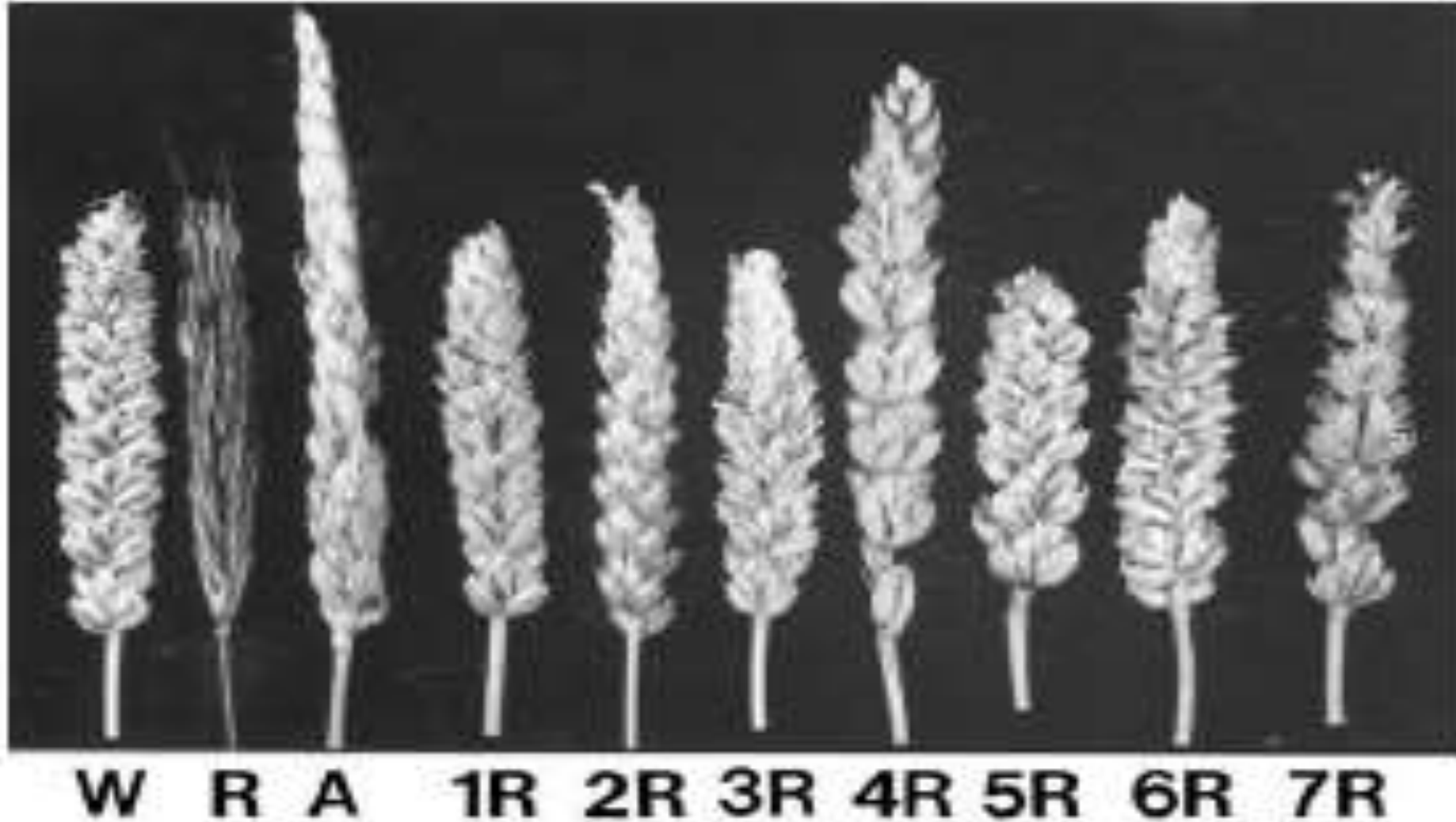
1. Gross morphology
2. Karyotype of the added chromosomes
3. Meiotic chromosome pairing in F1 plants obtained by intercrossing two DAALs
4. Isozyme analysis, and
5. By genetic tests

1. Morphological identification

MAALs and DAALs can be identified morphologically because

- The extra added chromosome(s) modifies the vegetative and spike morphological features.
- Alterations may be qualitative and quantitative.
- Alterations are due to interaction between genes of recipient and donor parents.
- Each chromosome has a specific effect on plant morphology and fertility.
- The addition of single chromosome (MAAL) from wild diploid wheat to tetraploid wheat alters morphological features of plants more drastically than those recorded in MAALs of hexaploid wheat.
- The addition of a alien chromosome to a diploid chromosome complement modifies the morphological features in the same way as observed in autotrisomics

Based on morphological features of spikes, 7 DAALs can easily be identified and distinguished in wheat-rye disomic addition lines

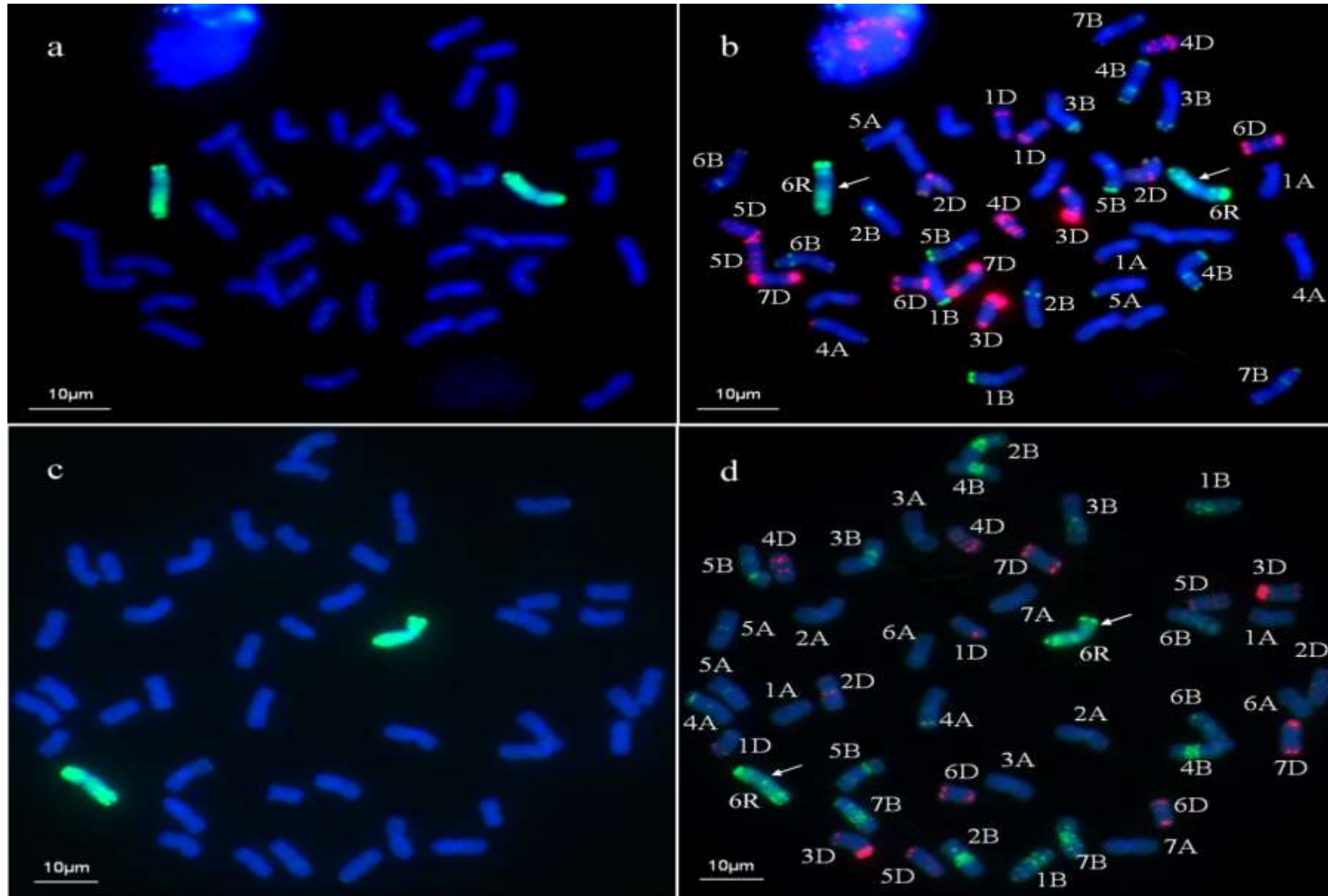


Each disomic wheat-rye addition expresses diagnostic morphological traits

Spikes of “Chinese Spring” wheat (W), “Imperial” rye (R), amphiploid (A), and seven disomic ($2n = 42W + 2R$) addition lines. (Courtesy of B. Friebe.)

• 2.CYTOLOGICAL IDENTIFICATION

- Chromosome morphology (total length, relative length, arm ratio) , Cytological chromosome markers



3. Meiosis in alien addition lines

- It is expected that a pair of alien chromosomes in DAALs should form a bivalent at metaphase-I.
- An alien chromosome generally remains as an univalent in MAALs and the chromosomes from the recipient parent synapse regularly. However, a low frequency of trivalent formation was occasionally seen

4. Intercrossing disomic alien addition lines

- Meiotic chromosome pairing in F1 hybrids is analyzed
- Alien addition lines carrying the same two chromosomes generally form a bivalent, the occurrence of two univalents indicates the DAALs are different

5. Biochemical identification

isozyme banding patterns are useful in identifying alien addition lines of wheat-rye, wheat-barley addition line.

- BREEDING BEHAVIOR OF ALIEN ADDITION LINES

- 1. Monosomic Alien Addition Lines

- univalent at meiotic metaphase-I.
- during gametogenesis, it can be eliminated or included in one of the gametes.

- 2. Disomic Alien Addition Lines

- DAALs are expected to breed true.
- Differences in meiotic cycles between recipient and donor chromosomes lead to the elimination of donor chromosomes.
- For example:
 - in wheat–rye DAALs, the rye chromosomes fail to function normally in the wheat nucleus, and the **tendency for asynapsis** may be due to the **absolute homozygosity of the rye chromosomes achieved during back crossing and selfing**.

- Fertility in alien addition lines

- Seed fertility in MAALs and DAALs is influenced by

1. The ploidy level of the recipient parent,

Ex. Seed set in MAALs and DAALs at the 6x level is expected to be better than at the 4x and 2x levels

➤ **The extra chromosome cause more imbalance at 2X and 4X than 6X.**

2. The genomic affinity between the recipient and donor species,

3. The genetic constitution of the added chromosomes.

• APPLICATIONS:

1. To transfer the chromosome carrying a desirable gene from one variety spices to another.
2. MAALs are powerful tools for elucidating genome structure .
3. MAALs can be used to study the introgression mechanism and the pairing status of homologous chromosomes.
4. The best procedure is to introduce the complete monosomic sets into the best commercial varieties.

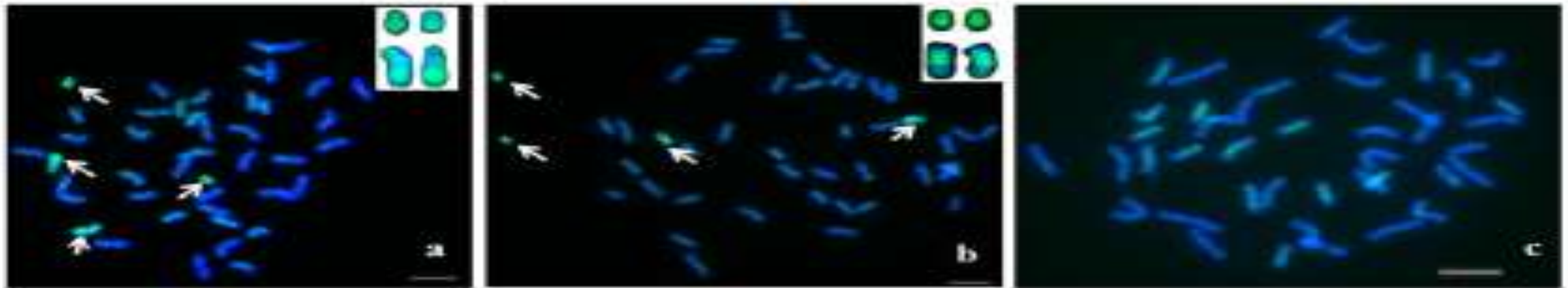
DRAWBACKS:

1. Genocidal effect under certain genotypes.
2. Imbalance produced in wheat genotype ultimately lose alien chromosome and return to euploid stage.

Microdissection and Chromosome Painting of the Alien Chromosome in an Addition Line of Wheat - *Thinopyrum intermedium*

Chuanliang Deng^{1,2}⊗, Lili Bai^{1,3}⊗, Shulan Fu¹⊗, Weibo Yin¹, Yingxin Zhang^{1,3}, Yuhong Chen¹, Richard R.-C. Wang⁴, Xiangqi Zhang¹, Fangpu Han^{1*}, Zanmin Hu^{1*}

Received March 11, 2013; Accepted July 11, 2013; Published August 14, 2013



Using *Thinopyrum intermedium* genomic DNA as the probe and “Chinese Spring” genomic DNA as the competitor DNA, the alien chromosomes in TAI-27 (wheat addition line) were identified (Figure a). The results showed that strong hybridization signals were uniformly distributed on two pair of chromosomes, of which one pair of chromosomes were the smallest in TAI-27.

Thank You

