

**Wheat Chromosome  
Engineering and Breeding**

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# Chromosome Engineering

- A process to transfer favorable alleles through **inter-specific hybridization** and interchange of chromatin using **aneupolids**

# Aneuploids?

- Individuals having chromosome numbers other than an exact multiple of **the basic chromosome set**.
- **A basic chromosome set** contains all chromosomes in **a genome**.
- **A genome** is defined as **the basic chromosome set** that contains all the genetic information needed to produce an organism, denoted by a  $x$ .

# Review Concepts

**n:** gametic (haploid) chromosome number.  $n = 3x = 21$  (wheat)

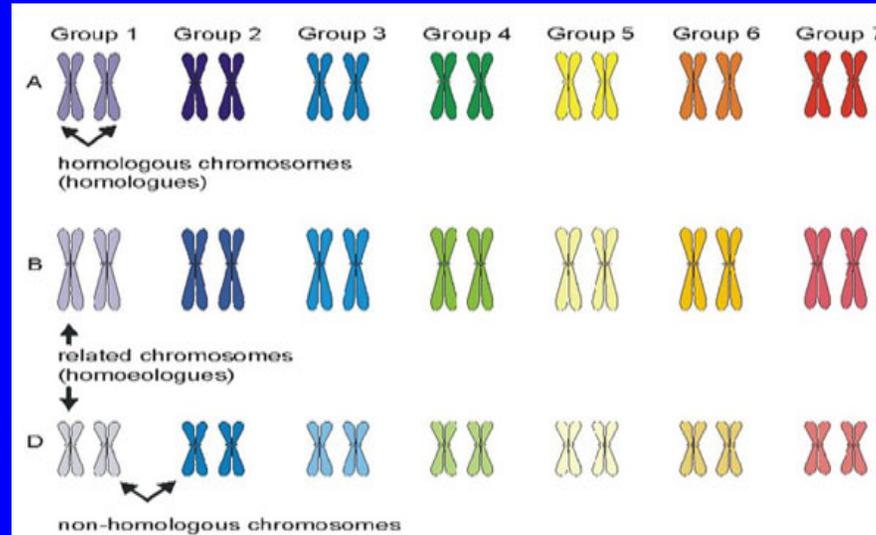
**2n:** disomic (somatic) chromosome number.

wheat:  $2n = 6x = 42$ ; Barley:  $2n = 2x = 14$ ; Soybean:  $2n = 2x = 20$ ; Maize:  $2n = 2x = 20$

# Outline of current lecture

- **Types of aneuploids in common wheat**
- **Application of aneuploids in wheat genetic and mapping studies**
- **Application of aneuploids in wheat breeding**

# Genetic Features



- Hexaploid wheat has homoeologous genomes A, B, D derived from diploid species (AA- *T. urartu*, BB- *Ae. Speltoides*, DD- *Ae. Squarrosa*)
- Most of the genes have three homoeologous loci, which can functionally compensate for one another.
- The homoeologous genomes can tolerate loss or addition of chromosomes.
- Complete sets of aneuploids of Chinese Spring and other wheat varieties are available.

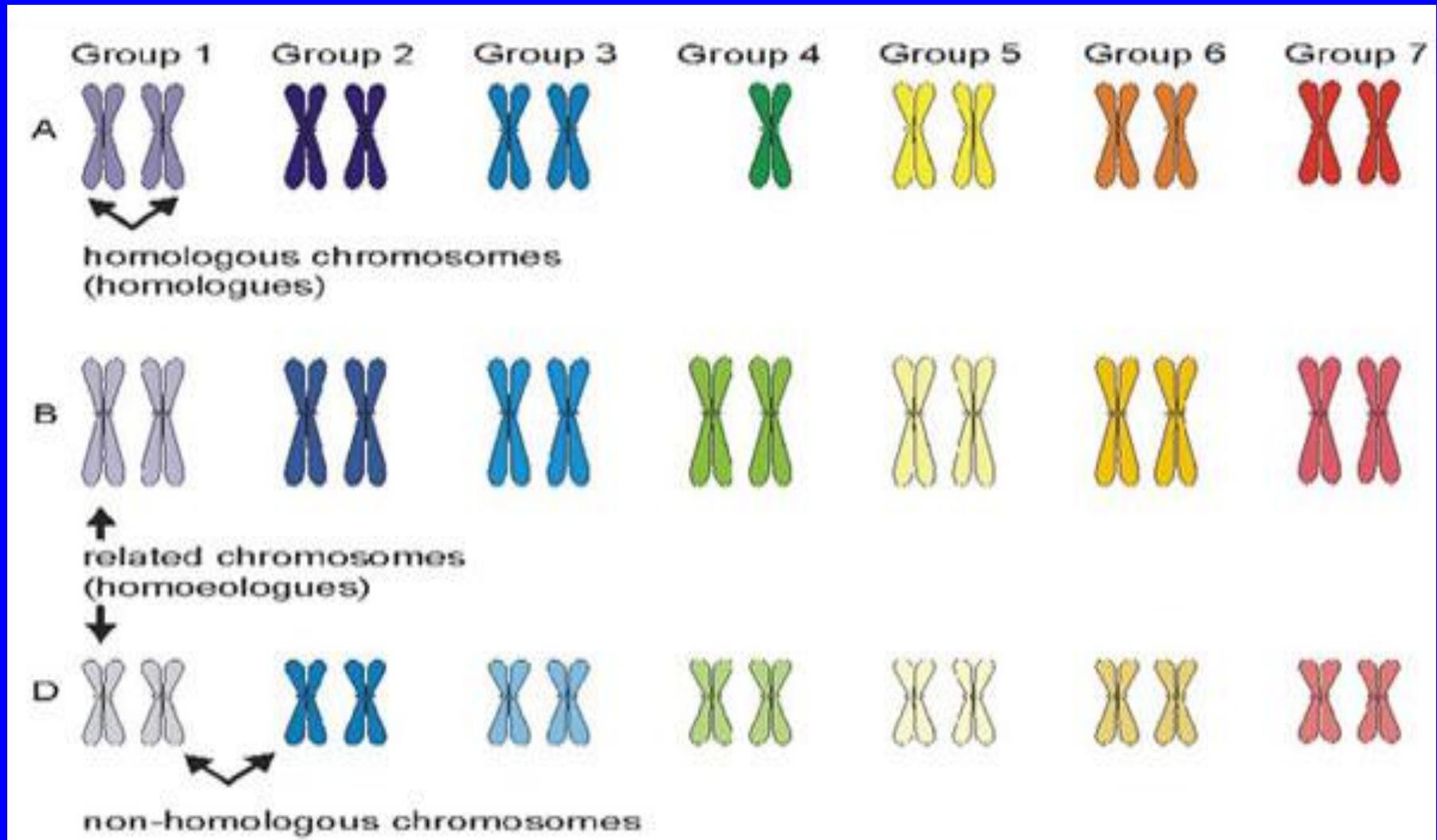
# Wheat Aneuploid Series

- Monosomic lines (Sears, 1954)
- Nullisomic lines (Sears, 1954; Xue et al., 1990 )
- Nulli-tetrasomic lines ( Sears, 1954).
- Ditelosomic lines (Sears and Sears, 1978).
- Deletion stocks ( Endo, 1978)

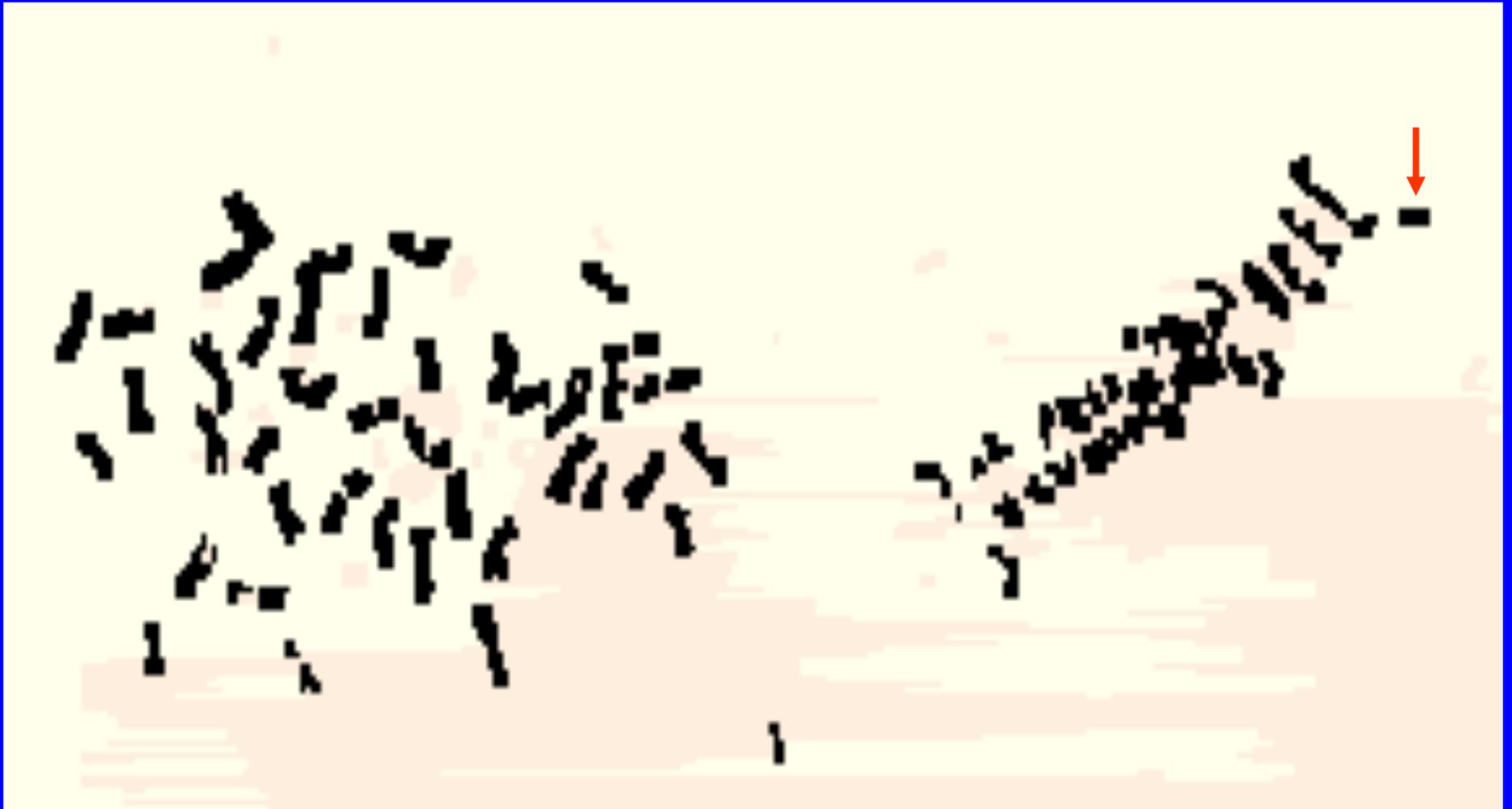
# Monosomics ( $2n-1$ ): a set of 21

- An individual **lacks** one of a pair of chromosomes from the normal diploid (disomic) complement ( $20 \text{ II} + 1 \text{ I}$  or  $41$ ).
- Plants look similar to the disomics and fertility is close to normal.
- Gametes are transmitted at a different rate through male and female.

# Monosomy 4A (2n-1) - 4AM



# Monosomic



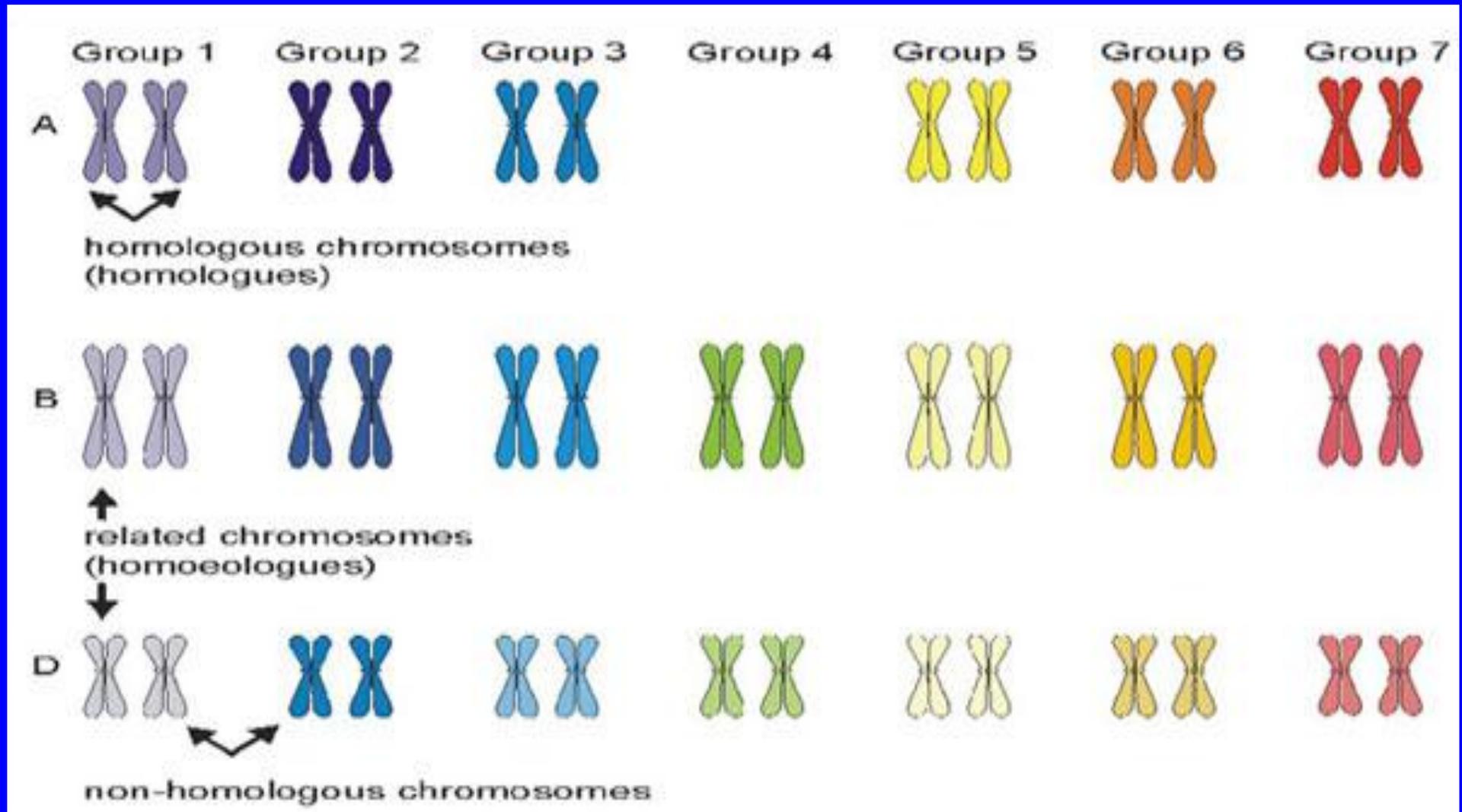
## Expected transmission of the monosomics in *Triticum aestivum*

	Male Gamete	
Female Gamete	n (21) 96%	n-1 (20 ) 4%
n (21 ) 25%	Disomic 2n (42) 24%	Monosomic 2n-1 (41) 1%
n-1 (20) 75%	Monosomic 2n-1 (41) 72 %	Nullisomic 2n-2 (40) 3%

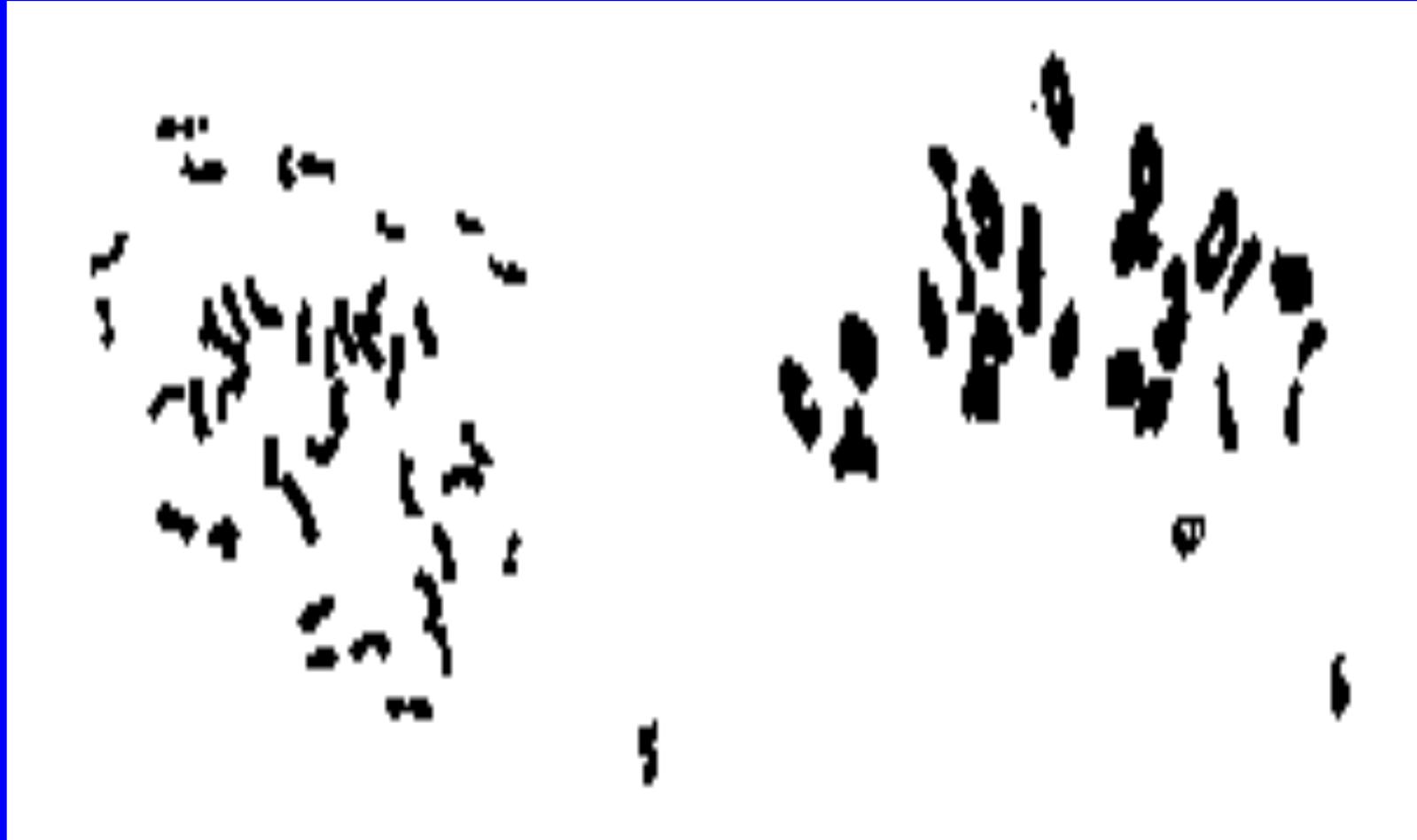
# Nullisomics ( $2n-2$ ): a set of 21

- An individual lacks of one pair of chromosomes from the normal diploid complement (20II or 40)
- Plants distinguishable by morphological features (vigor and size)
- Gametes transmitted via same rate in female and male

# Nullisomy 4A ( $2n-2$ ) – 4AN

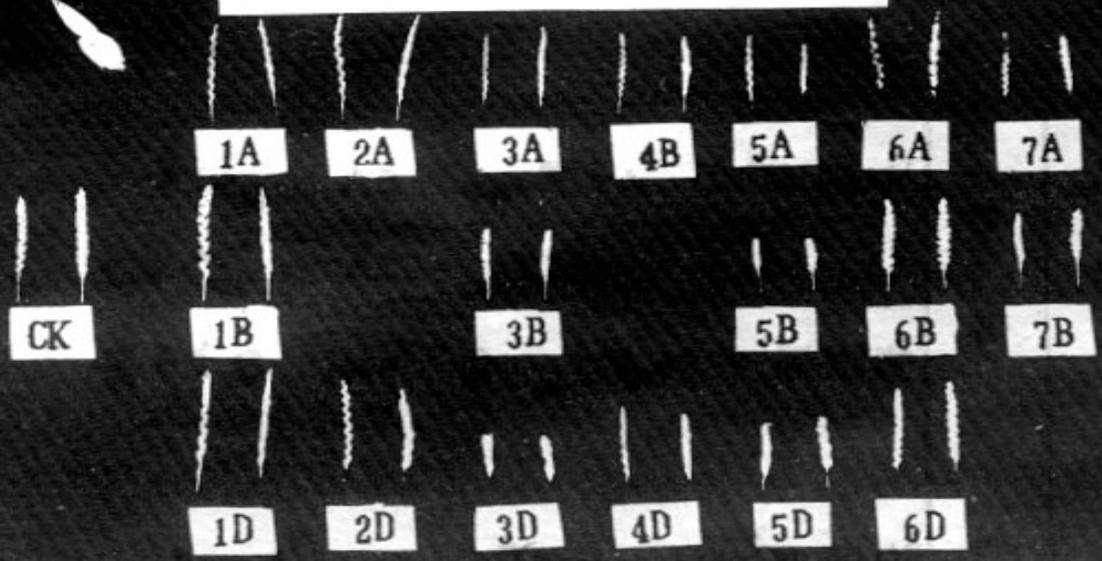


# Nullisomic

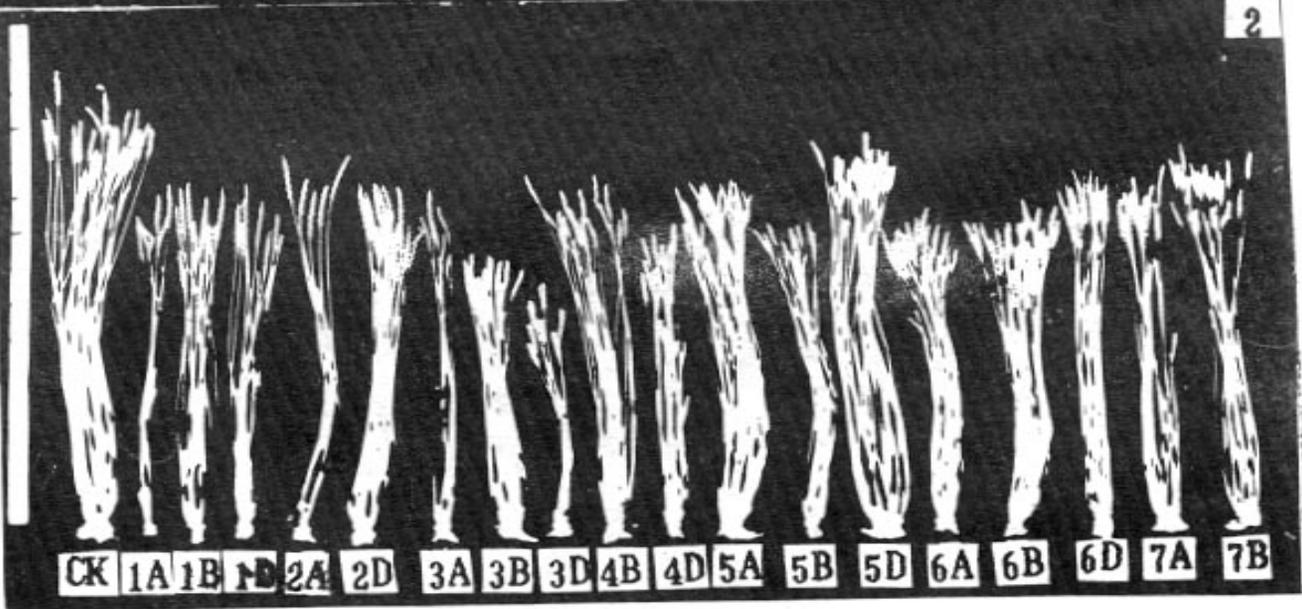


阿勃稳定自交结实的缺体系统

1



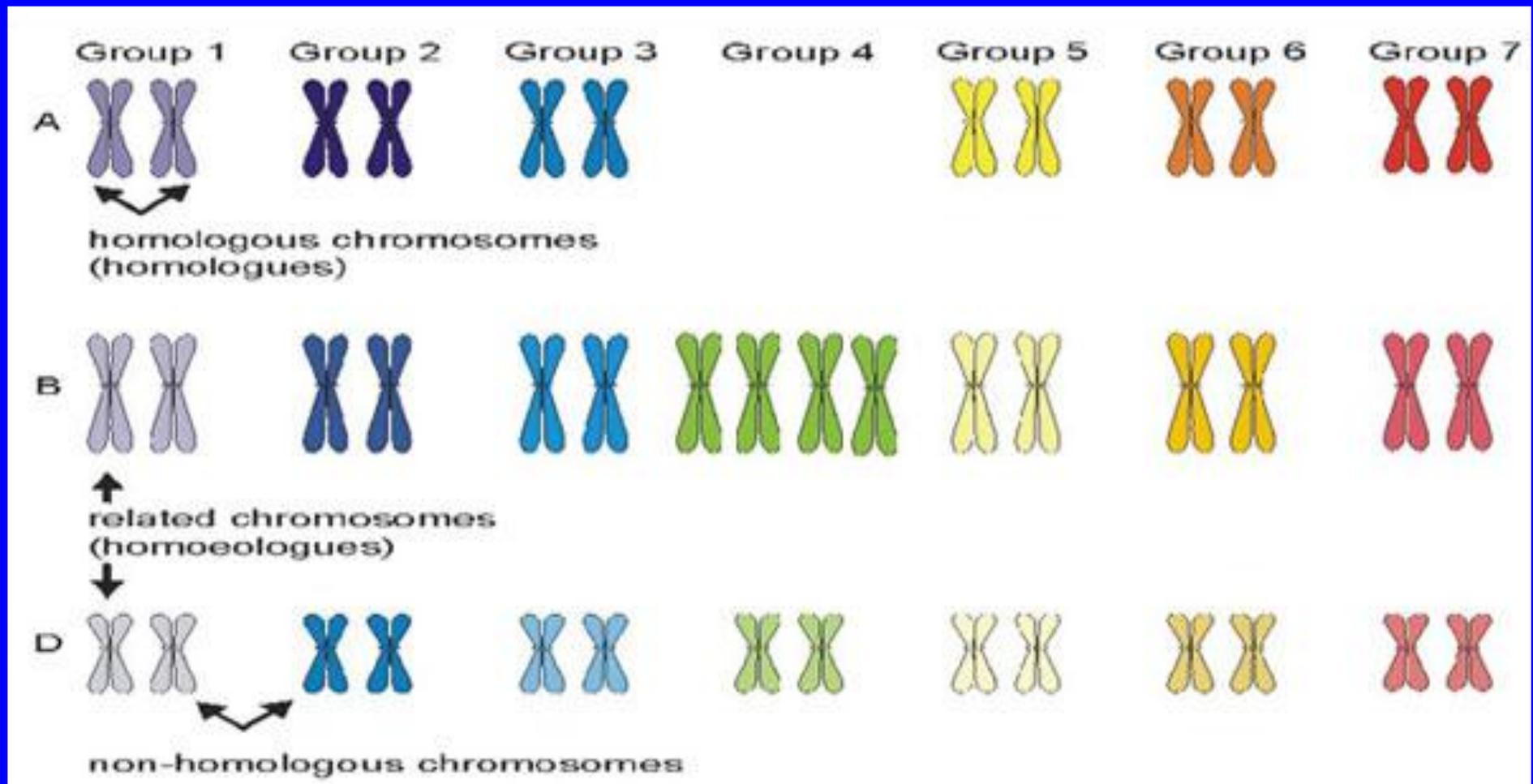
2



# Nulli-tetrasomics: a set of 42

- An individual lacks one pair of chromosomes but have a doubled pair of chromosomes from the homoeologous group (19 II + 1IV, or 42)
- Plants look similar to disomics and fertility is close to normal
- Gametes transmitted via similar rate in female and male

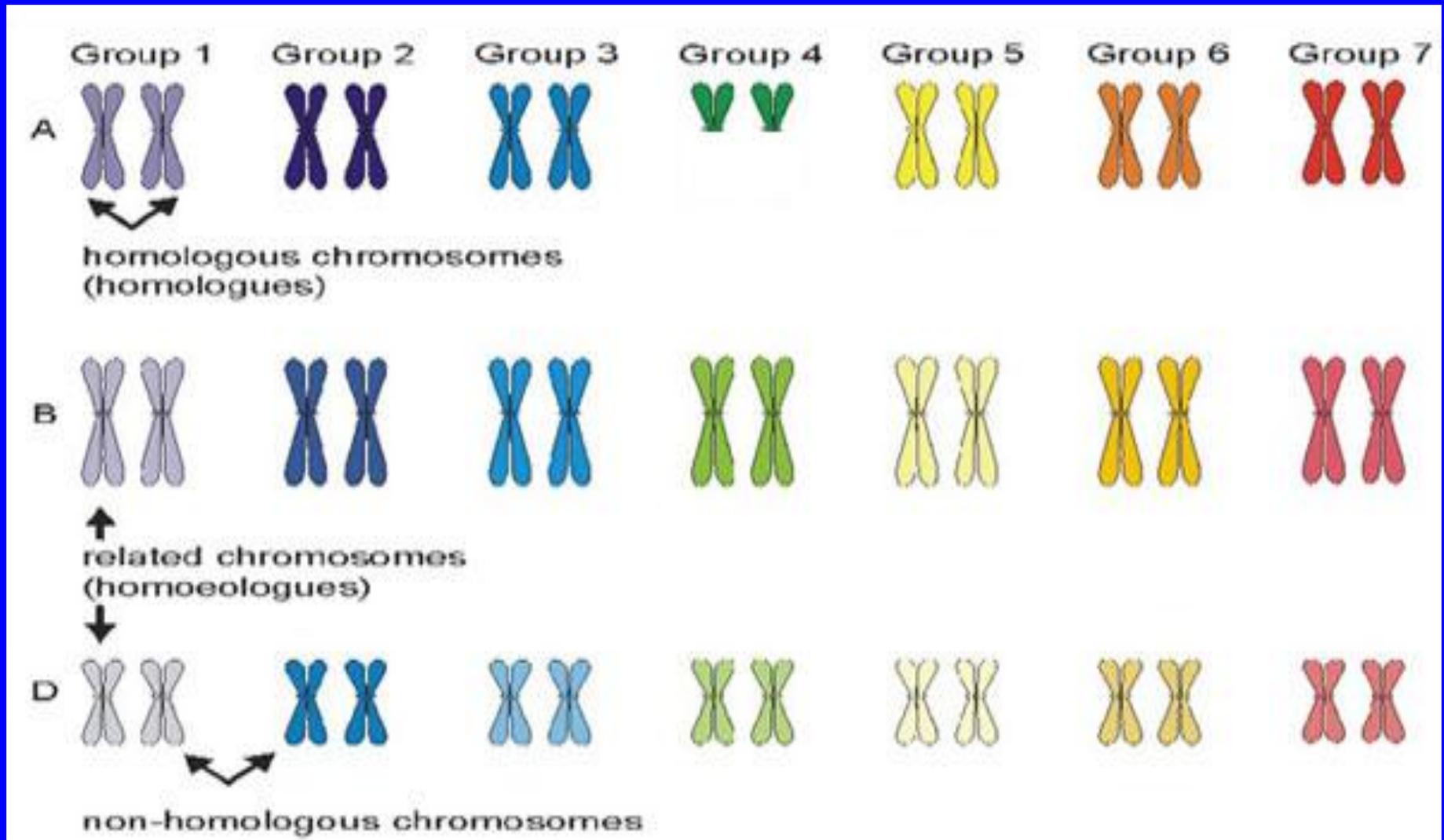
# Nullisomy4ATetrasomy4B - N4AT4B



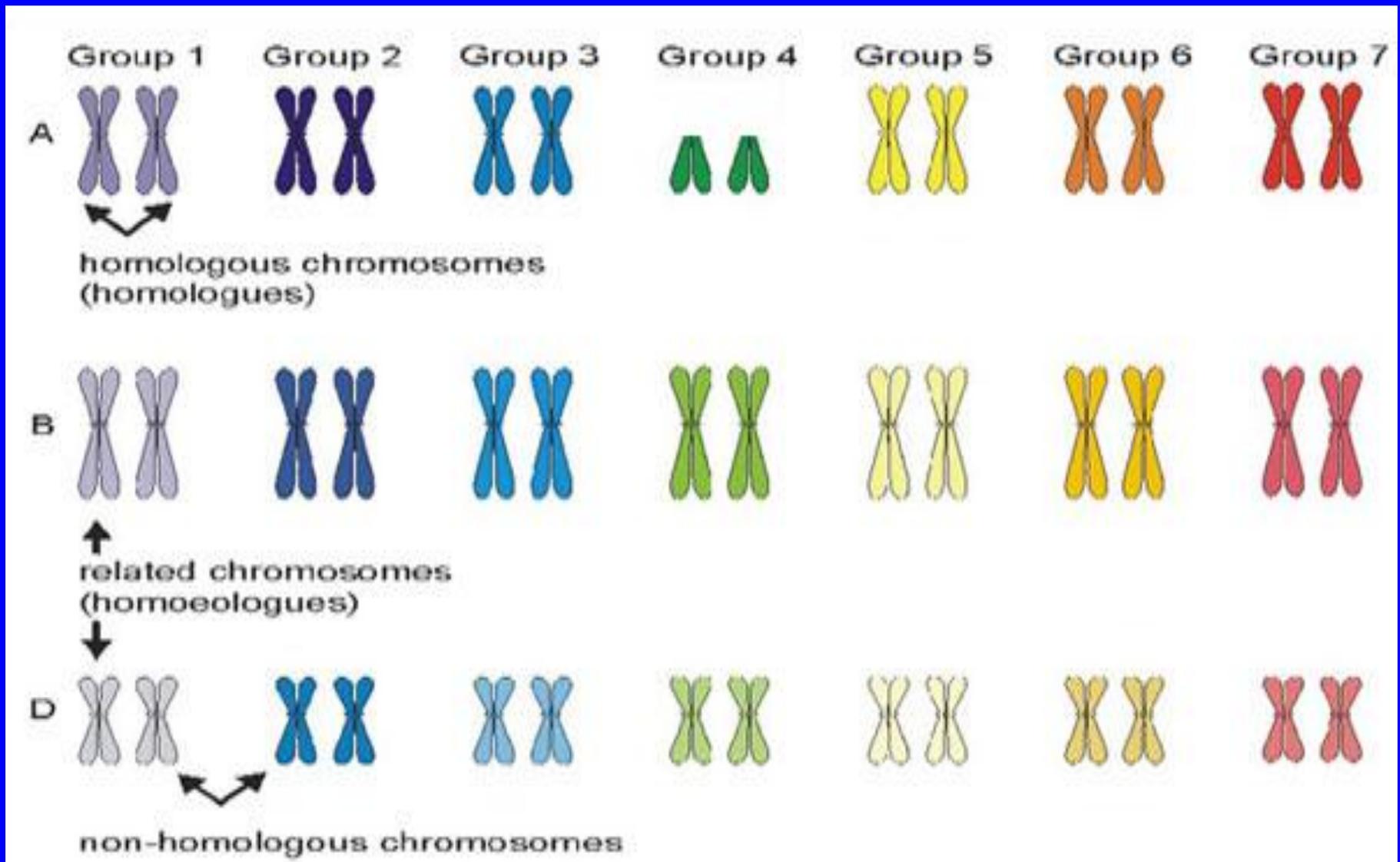
# Ditelosomics: a set of 42

- An individual lacks **a pair of chromosome arms** from the normal diploid complement
- Plants look similar to disomics and fertility is close to normal
- Gametes transmitted via same rate in female and male

# Ditelosomy 4AL – 4AL



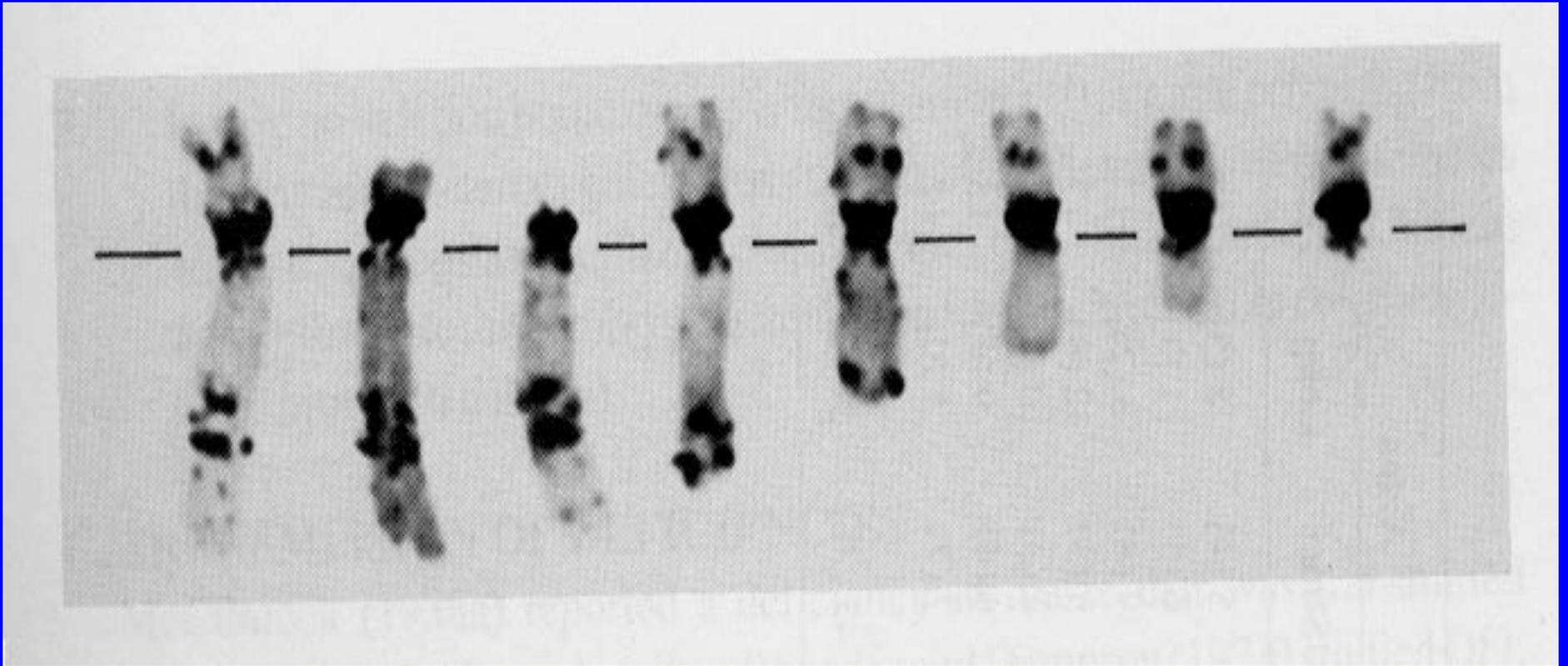
# Ditelosomy 4AS – 4AS



# Deletion stocks: unlimited

- An individual lacking **a segment of a chromosome** from the normal diploid complement
- Fertility is based on the location of deletions
- Mainly used in physical mapping

## 5B Deletion stocks



(Endo, T.R. 1990. Jpn. J. Genet. 65: 135-152)

# Genetic Study

Locate genes of interest:

on specific chromosome

on specific chromosome arm

on specific chromosome

segment

# Monosomic analysis – F1

	Male gamete
Female gamete	n (21)
n (21 chr., 25%)	Disomic ( $2n = 42$ , 25%)
n-1 (20 chr., 75%)	Monosomic ( $2n-1 = 41$ , 75%)

# Expected transmission of the monosomics - F2

Female Gamete	Male Gamete	
		n (21) 96%
n (21) 25%	Disomic 2n (42) 24%	Monosomic 2n-1 (41) 1%
n-1 (20) 75%	Monosomic 2n-1 (41) 72 %	Nullisomic 2n-2 (40) 3%

# Locating Recessive Genes (1A is gene carrier)

Female		Male
(Awnless)		(Awn)
$20\text{II} + 1\text{I}_{1A}$	X	$20\text{II} + 1\text{II}_{1aa}$
	↓	
F <sub>1</sub>	75%	$20\text{II} + 1\text{I}_{1a}$
	25%	$20\text{II} + 1\text{II}_{1Aa}$

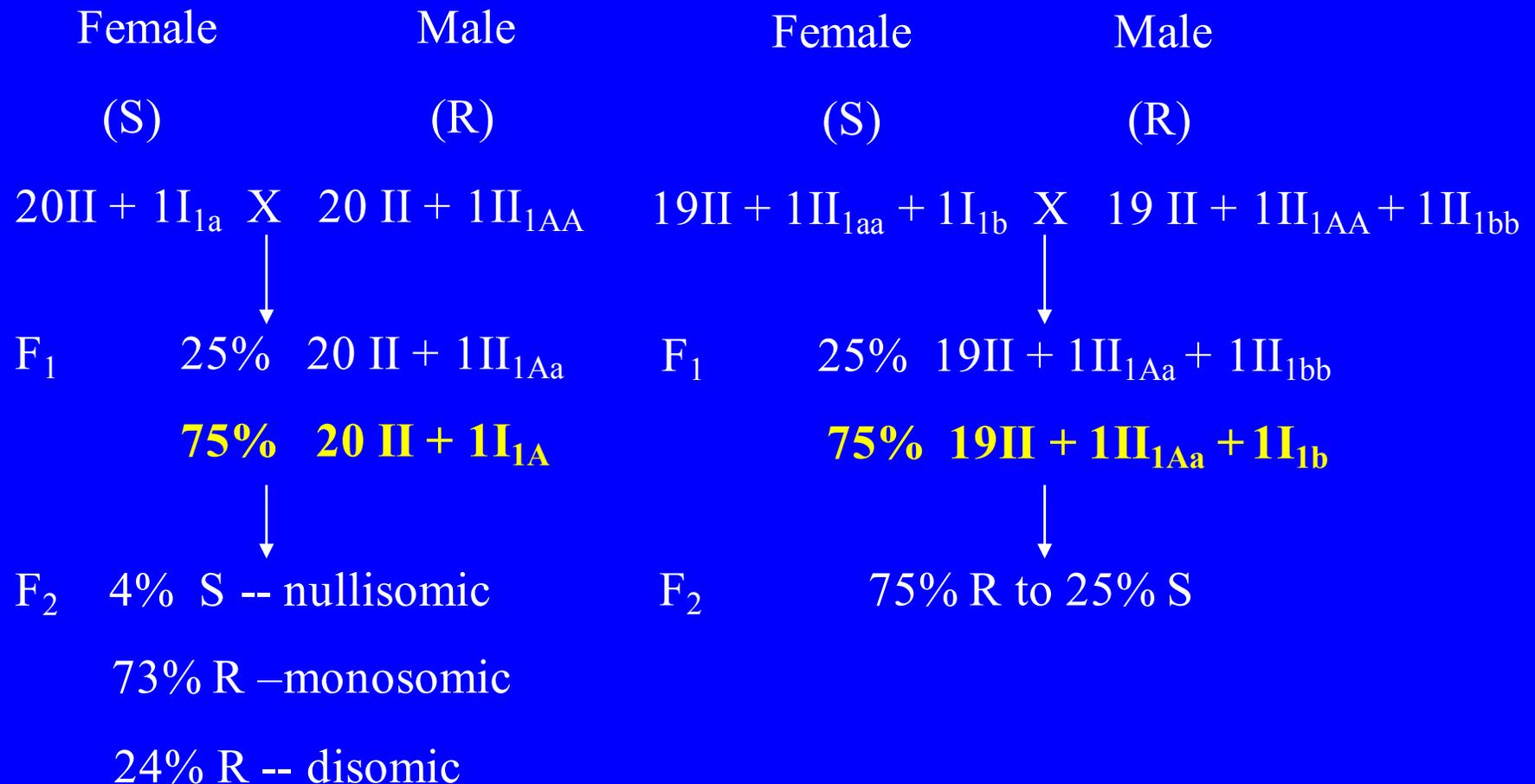
3(Awn and monosomic)

1(Awnless and disomic)

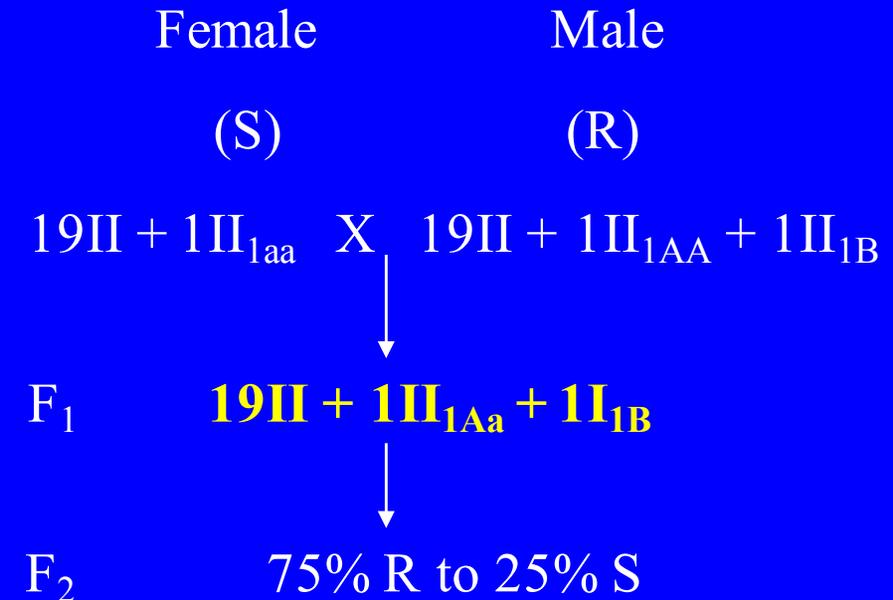
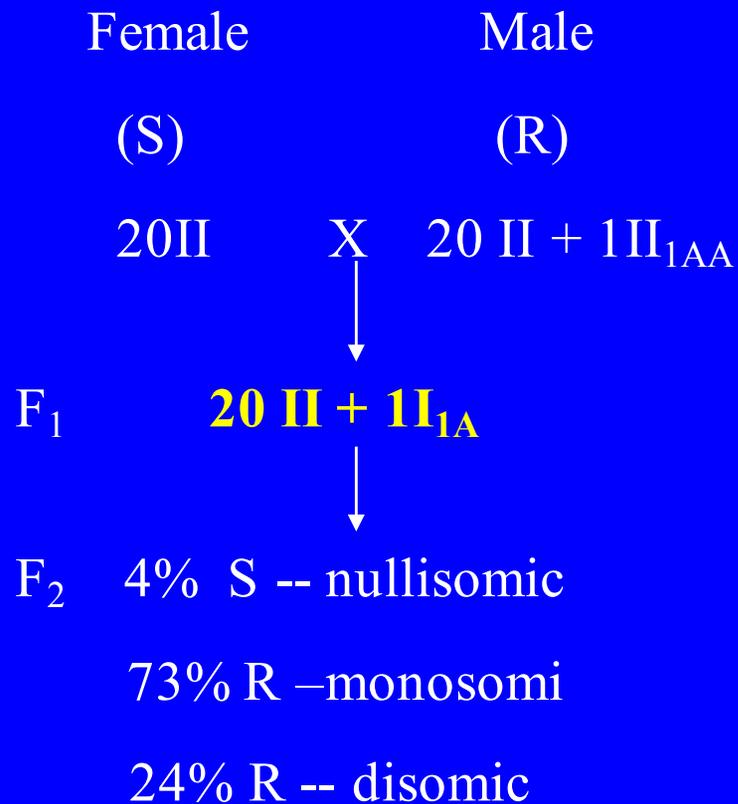
Female		Male
(Awnless)		(Awn)
$19\text{II} + 1\text{II}_{1AA} + 1\text{I}_{1b}$	X	$20\text{II} + 1\text{II}_{1aa}$
	↓	
F <sub>1</sub>	75%	$19\text{II} + 1\text{II}_{1Aa} + 1\text{I}_{1b}$
	25%	$19\text{II} + 1\text{II}_{1Aa} + 1\text{II}_{1bb}$

All plants are awnless

# Locating Dominant Genes (Monosomic Analysis)



# Locating Dominant Genes (Nullisomic Analysis)



**No cytogenetic work in F<sub>1</sub> !**

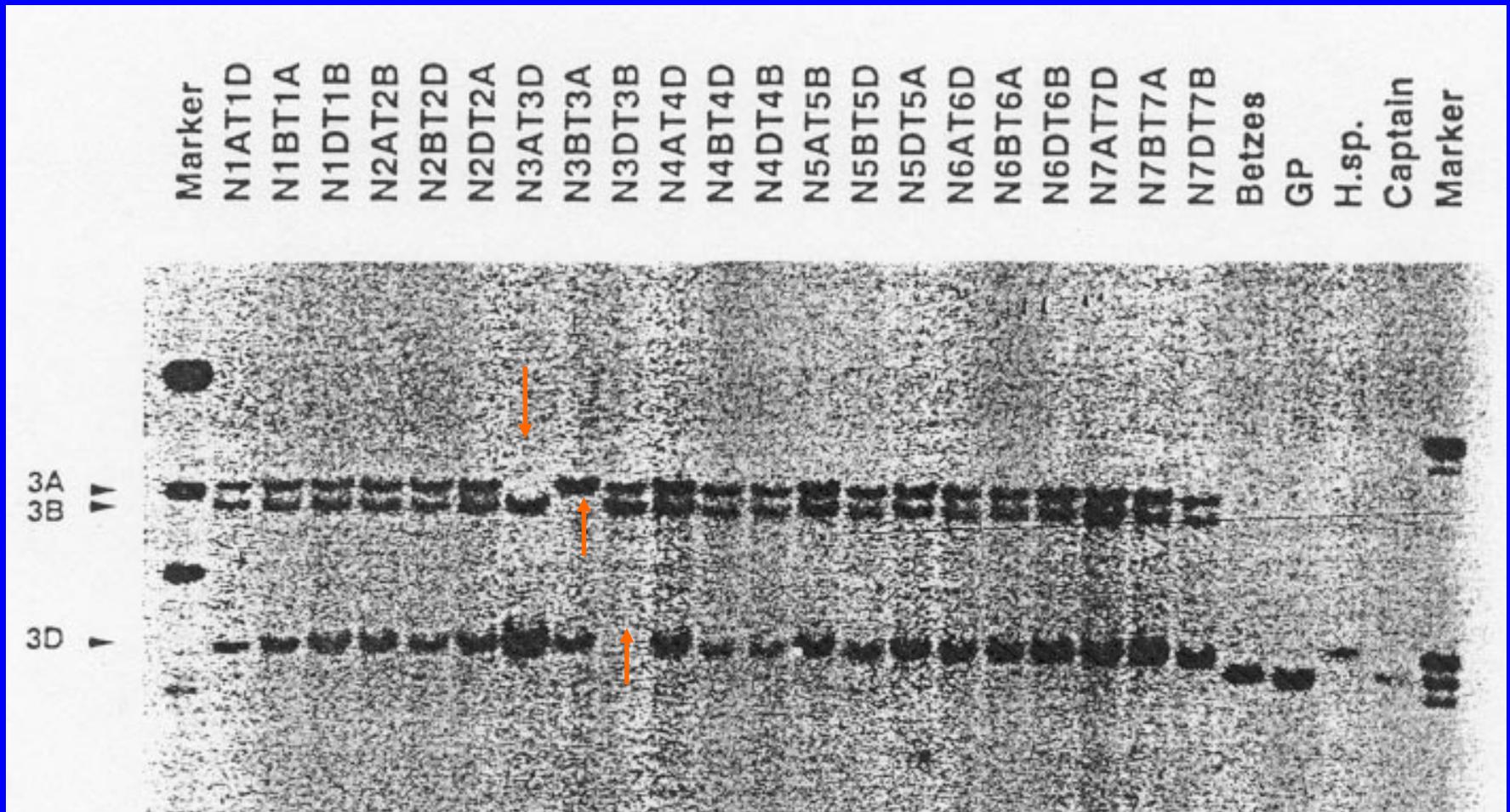
# Genome Mapping

Locate genes of interest or gene tightly

linked markers

- on specific chromosome
- on specific chromosome arm
- on specific chromosome segment

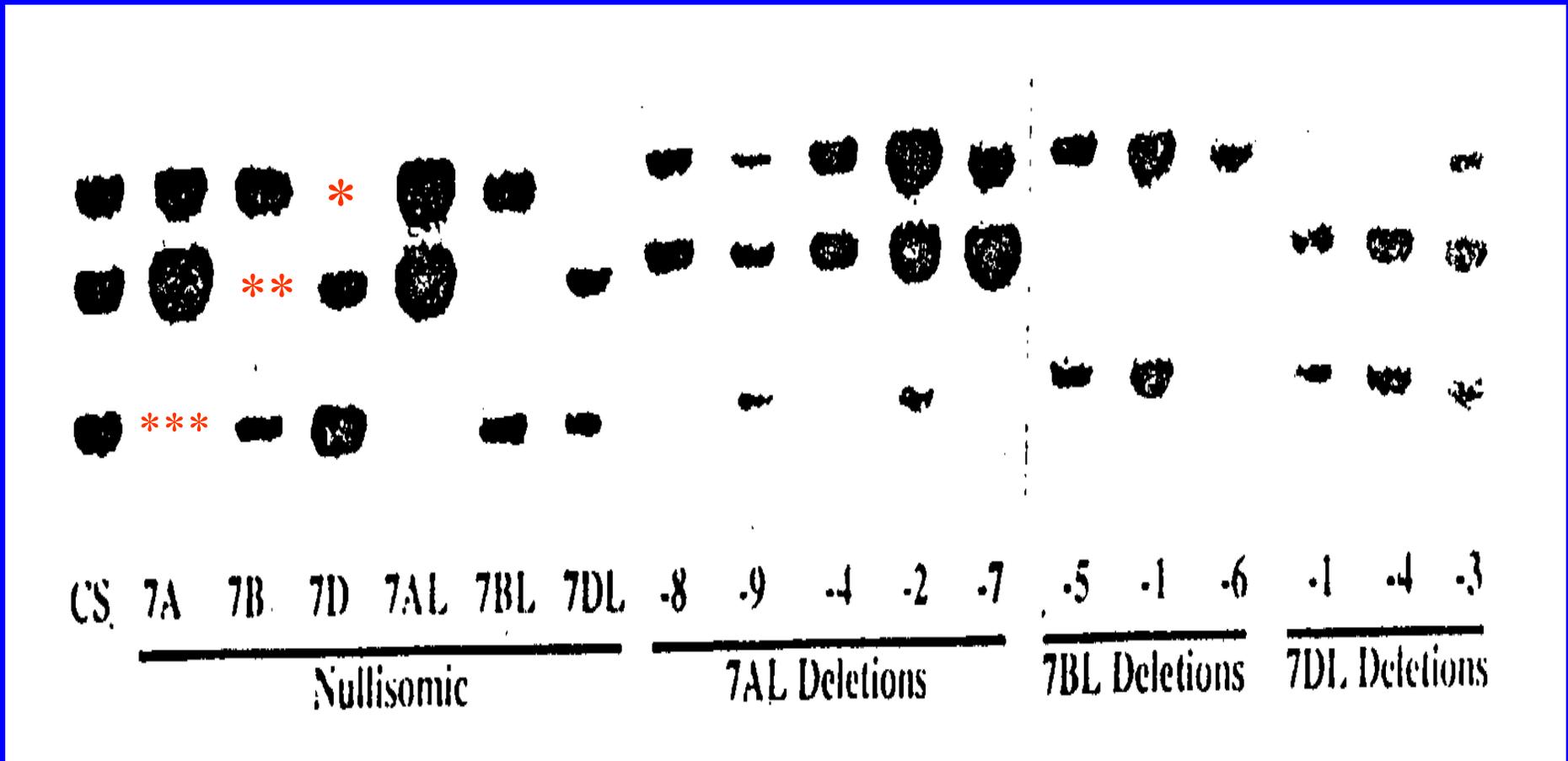
# Nulli-tetrasomic analysis - RFLP



( Devos KM and Gale MD. 1993. Theme:93-99)



# Deletion analysis – locate markers on chromosome segment



# Variety Improvement: interspecific hybridization

- Introgression of useful traits from alien species to common wheat
- Issues on interspecific crosses
  - sterile or partially sterile in  $F_1$
  - effective way to tag introduced chromosome or chromosome segments
  - Chromosome banding, genomic in situ hybridization, and molecular marker assisted selection

# Procedures for transferring useful genes

- Screening of donor populations
- **Producing hybrids**
- Chromosome doubling/backcrossing
- **Production and identification of alien addition/substitution lines**
- **Induction of recombination**
- **Screening/stabilizing recombinants/translocations**
- Gene tagging/Newer technologies

# Useful agronomic traits in alien species

## Disease resistance

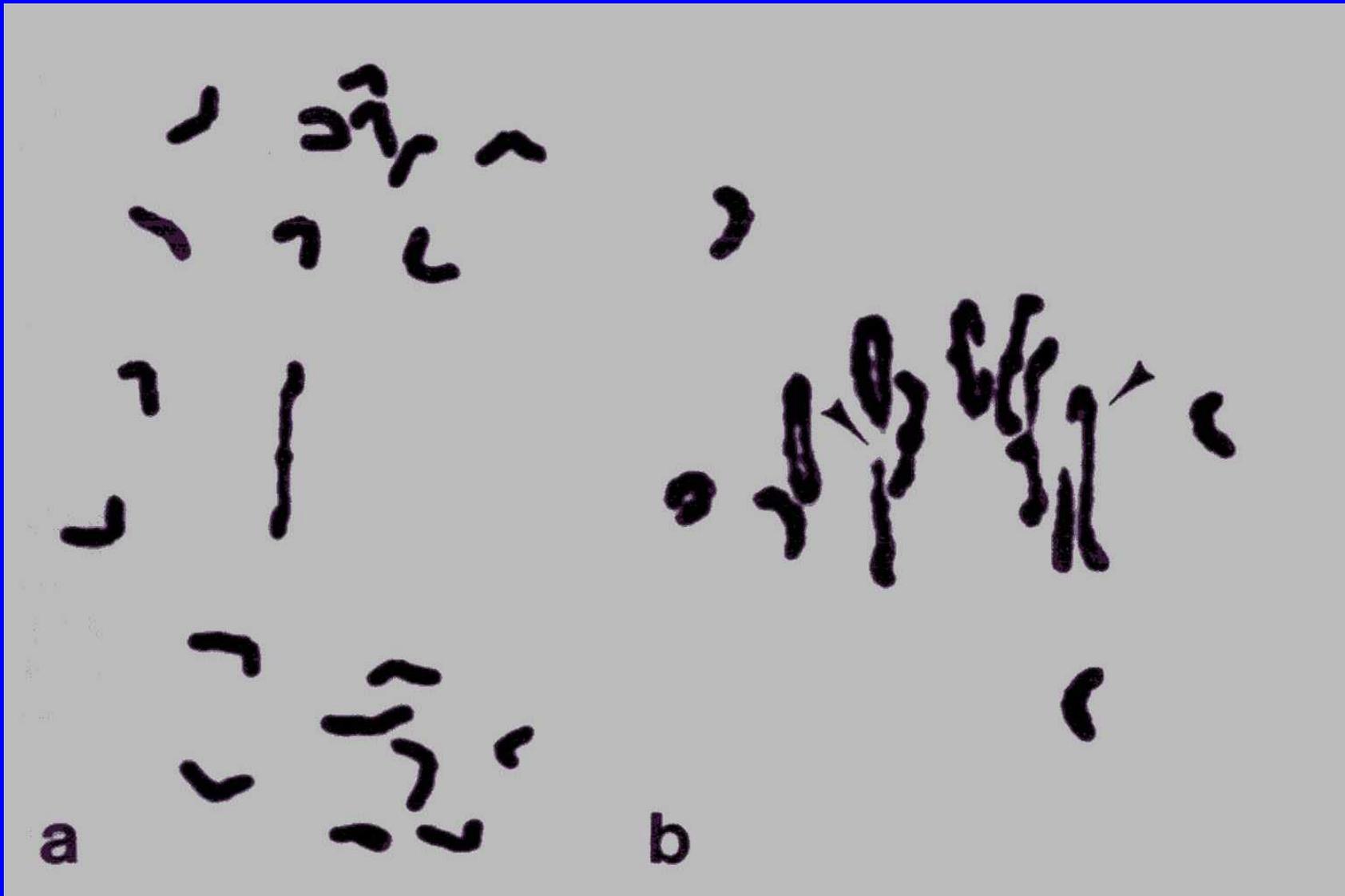
- Powdery mildew – *Ae. Markgrafii*, *Ae. Comosa*, *D. villosum*, *T. spelta*, *T. dicoccoides*, *T. macha*, *Ae. kotschii*
- Leaf rust – *Ae. Caudata*, *T. monococcum*, *T. tauschii*, *Th. Distinchum*
- Stem rust – *T. diccoides*, *T. tauschii*
- Yellow rust – *T. spelta*, *synthetic hexaploids*
- Karnal bunt – *triticales (4R, 6R)*, *T. monococcum*

# Method of Translocation Induction

- Tissue culture (BYDV) (Bank et al., 1995)
- Radiation (Lukaszewski, 1995)
- **The 5B system – induction of homeologous pairing (Sears, 1977)**

# The 5-B system in wheat

- The 5B system, a Ph gene (homoeologous pairing suppressor) is a genetic control which restricts chromosome pairing to homologs.
- When Ph is **removed**, or its activity is **suppressed**, not only do homoeologous chromosome pair but they also pair with the chromosome of related species and genera, making alien gene transfer possible (Sears, 1975, 1976).

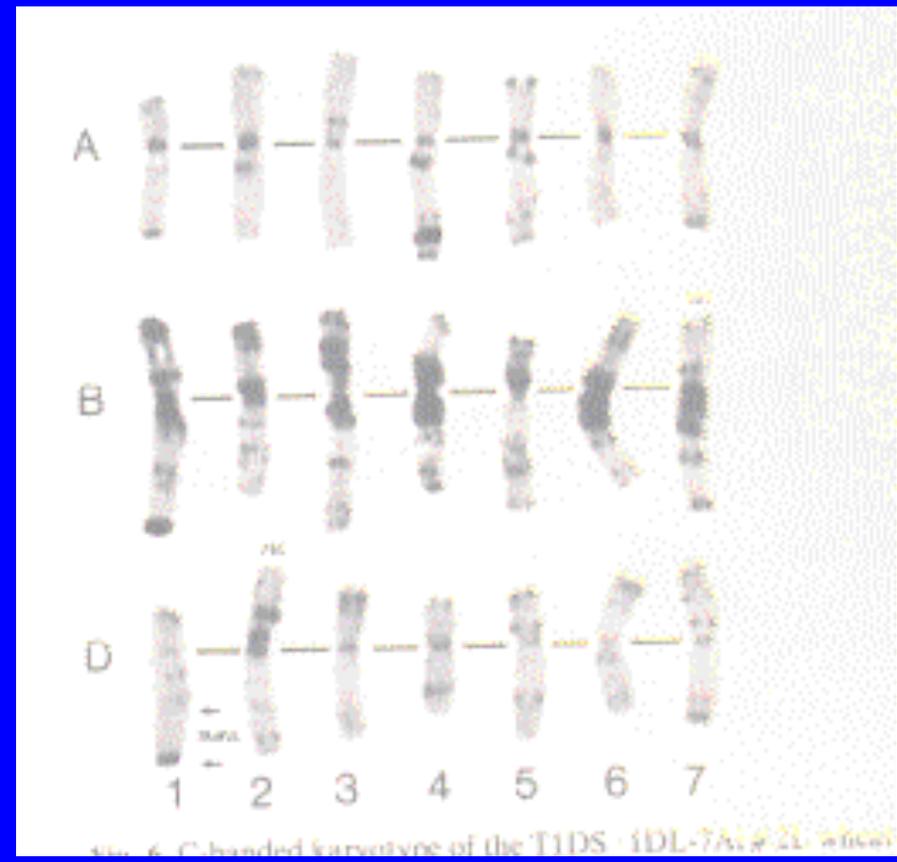
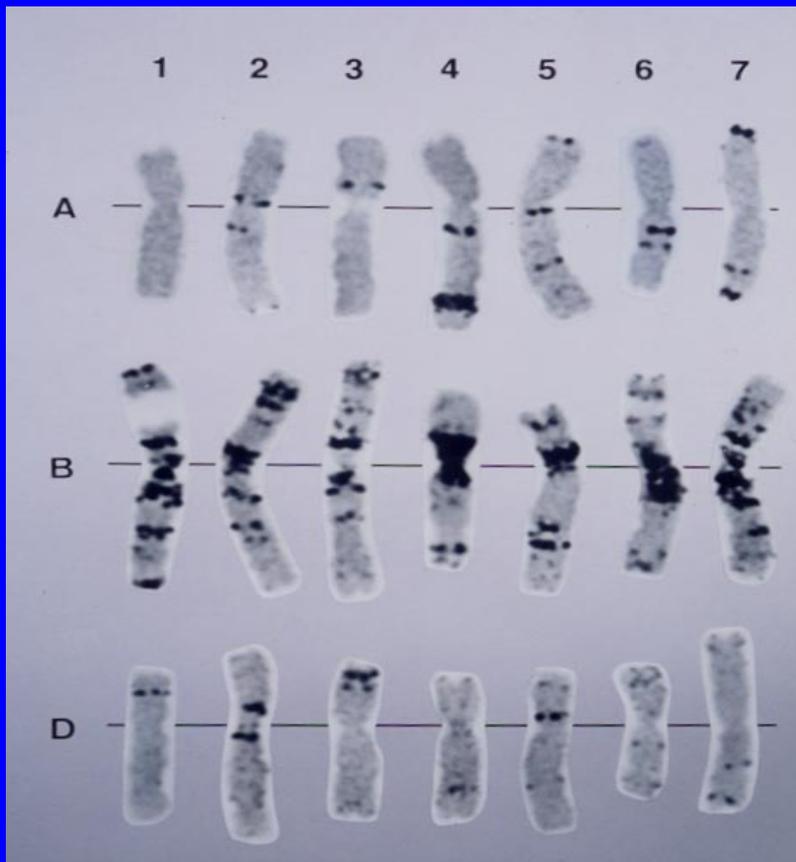


**Chromosome pairing in polyhaploids of bread wheat ( $2n = 21$ ) with (a) and without (b) *Ph* gene (from Jauhar *et al.*, 1991)**

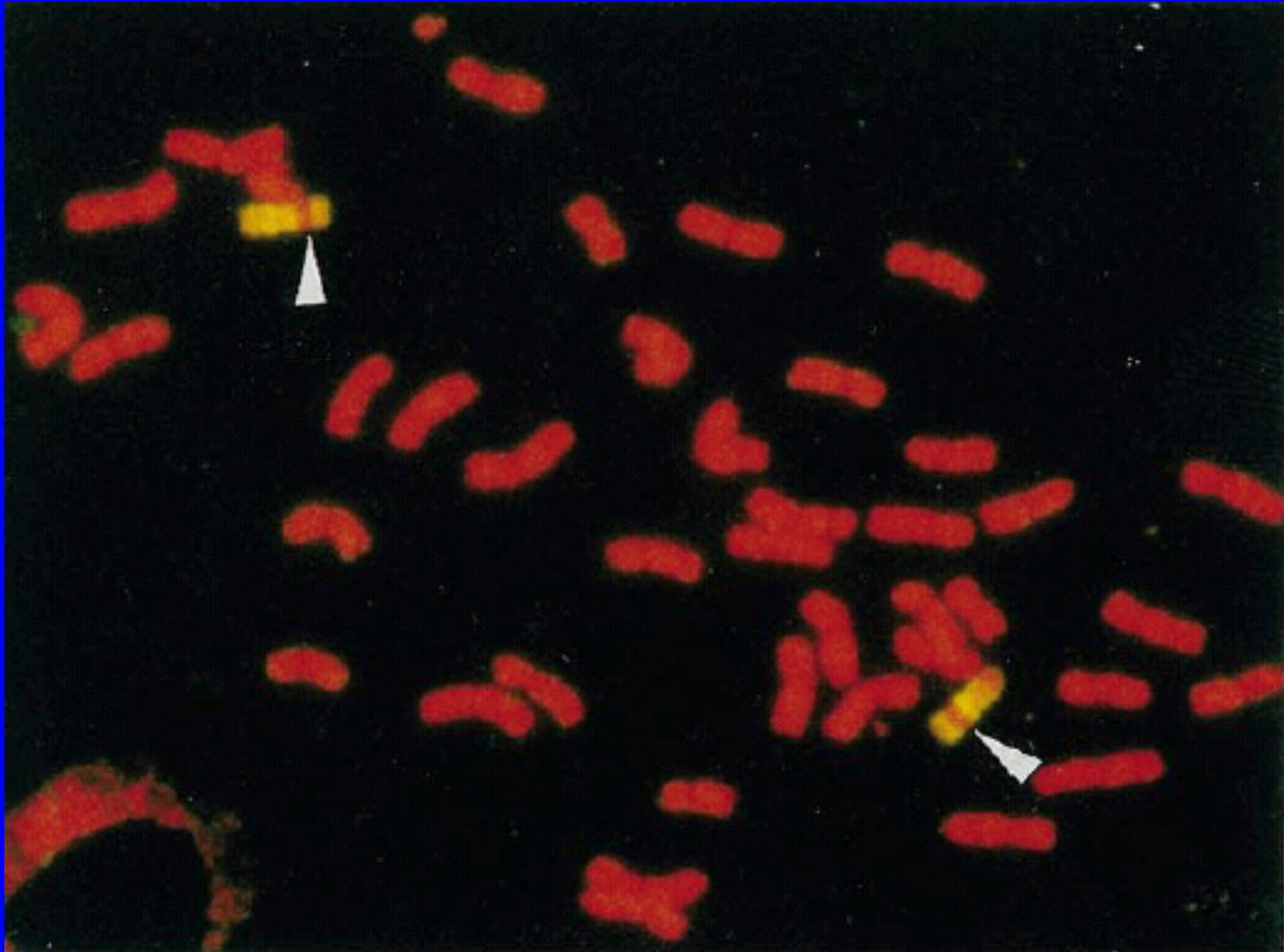
# Procedures for inducing homoeologous pairing

1. Monosomic or Nullisomic 5B x Alien species,  $F_1$  x adapted lines or variety;
2. Monosomic or Nullisomic 5B x Alien addition or substitution lines,  $F_1$  x adapted lines or variety.

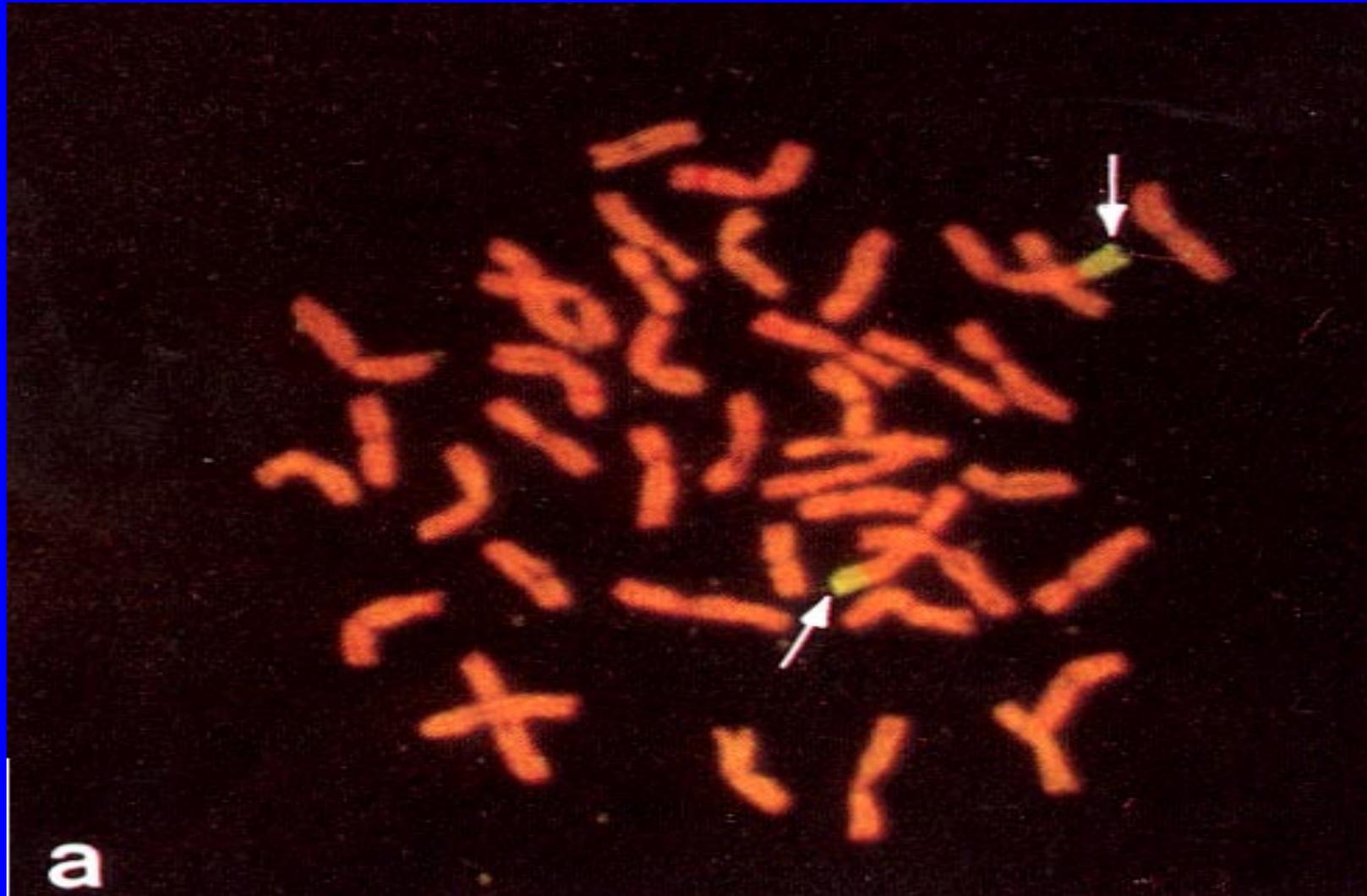
# Identification wheat-alien translocation lines



# Identification wheat-alien substitution lines



# Identification wheat-alien translocation lines



# Identification wheat-alien addition lines

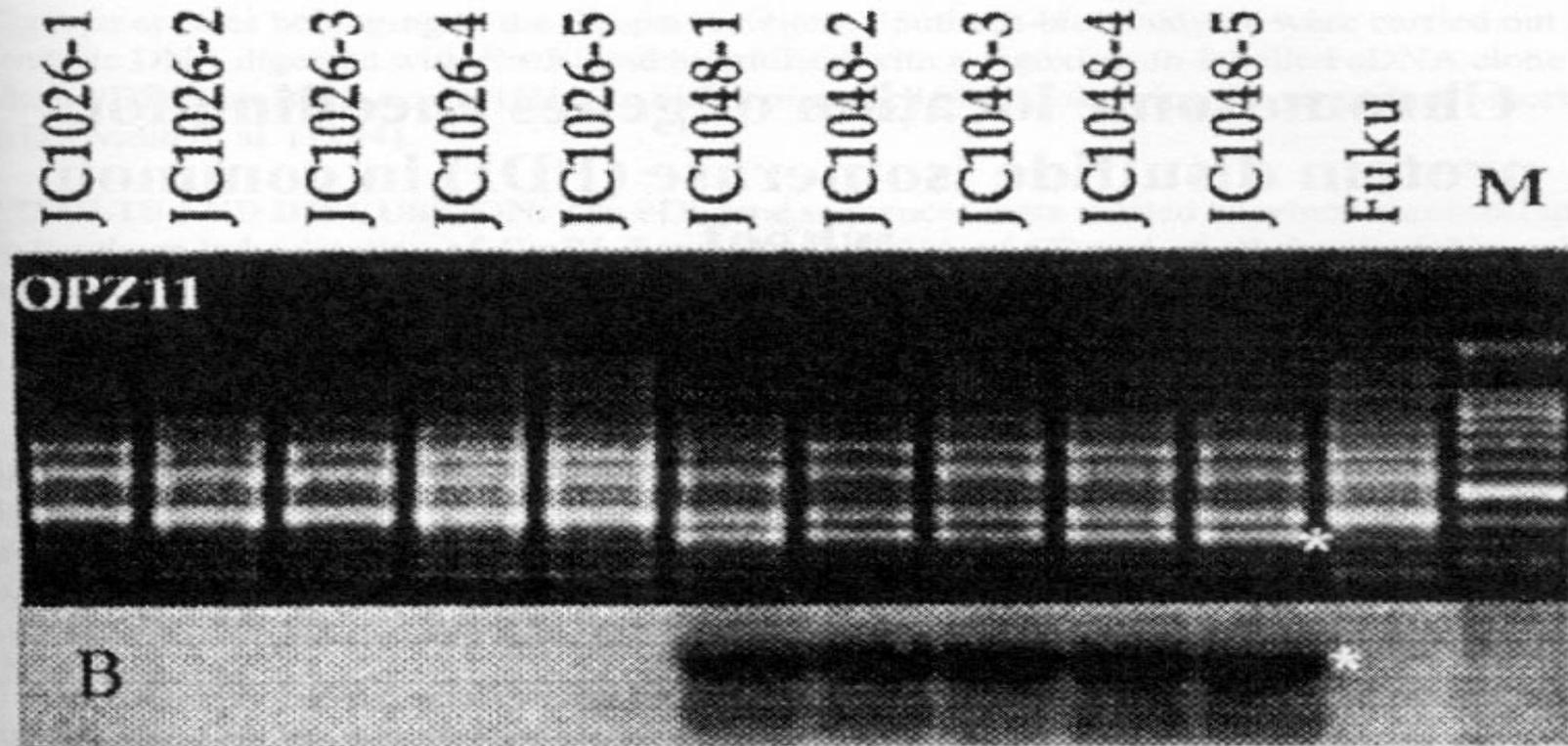


Figure 3 RAPD analysis in 10 individuals of two addition lines from JC1050. RAPD was performed by primer OPZ11(C), and the product was hybridized with OPZ11-350 (C1).

# Conclusion

- Aneuploids are unique in hexaploid wheat.
- Genome analysis in wheat has served as a model in other plant systems, and has made tremendous advances.
- Chromosome engineering will continue to make contributions to wheat improvement as new techniques become available.

# Aneuploid Analysis

## Practical considerations

- Availability of aneuploid stocks in your crop species
- Nature of genes: qualitative or quantitative; dominant or recessive