

Session-16

A monosomic maize plant contains one copy of one chromosome and two copies of each of the other nine chromosomes ($2n - 1 = 2x - 1 = 19$ chromosomes). There is a genetic system in maize, the r-XI system that generates a high frequency of monosomic and trisomic plants.

The r-XI Deficiency: The r-XI deficiency, a deficiency that includes the R locus on chromosome 10 in maize, was X-ray induced by L. J. Stadler (unpublished). The dominant allele of the R locus is necessary for anthocyanin production. Plants germinated from colorless kernels (that contain the r-XI deficiency) include monosomics and trisomics along with diploids. Clearly, a factor on chromosome 10 that is necessary for normal chromosome disjunction is deleted in the r-XI deficiency. Nearly all of the monosomics produced by the r-XI system have a chromosome missing from the entire plant. Each of the monosomic types is distinctively smaller but remarkably vigorous than its diploid siblings. Using the r-XI system several thousand maize monosomics that include monosomies for each of the 10 chromosomes were generated.

Some Characteristics of the r-XI System: Some of the characteristics of the r-XI deficiency are as follows:

- (1) It is submicroscopic (too small to be detected using the light microscope).
- (2) It is transmitted with a high efficiency through female gametes; however, pollen grains containing it do not function even though they appear to be morphologically normal.
- (3) It is due to nondisjunction at the second mitosis of the three embryo sac divisions (during embryo sac development).
- (4) The genetic background influences the activity of the r-XI system.

Application of r-XI System

It has also been used successfully to assign a biochemical marker (the benzoxazinless locus), a histone electrophoretic variant, and an isozyme locus of peroxidase to the respective chromosomes. Monosomics have been compared with diploid sibling plants in studies of gene dosage effects on factors controlling the level and composition of embryo lipids, the free fatty acid composition in leaves, and recombination. The system also can be an efficient and economical means for assigning cloned DNA fragments to specific chromosomes.

Alien-gene transfer through chromosome manipulations

The success of introducing alien variation into crop species from related species depends on the cytogenetic relations between the species. If there are no restrictions on chromosome pairing and recombination in species hybrids, a backcrossing programme can be used to obtain the desired gene transfers. However, when there is a failure of adequate chromosome pairing in species hybrids, techniques of chromosome manipulation have to be used to obtain alien gene transfers. In polyploid crop species it is possible to introduce appropriate single chromosomes of the alien species into the genotype of the recipient species, but the failure of the alien chromosome to become integrated into the genotype of the recipient species often leads to meiotic instability. The introduction of segments of alien chromosomes has been successful through the use of irradiation-induced translocations. The deletions/duplications that are a consequence of such translocations do limit the usefulness of this approach. In a number of allopolyploid crop species, regular bivalent pairing behaviour has been shown to be genetically controlled. By interfering with the genetic system controlling the diploid-like pairing it is possible to induce pairing between the alien chromosomes and its corresponding chromosomes in the crop species. Gene transfers based on this method involve exchanges between chromosomes of similar gene sequences.

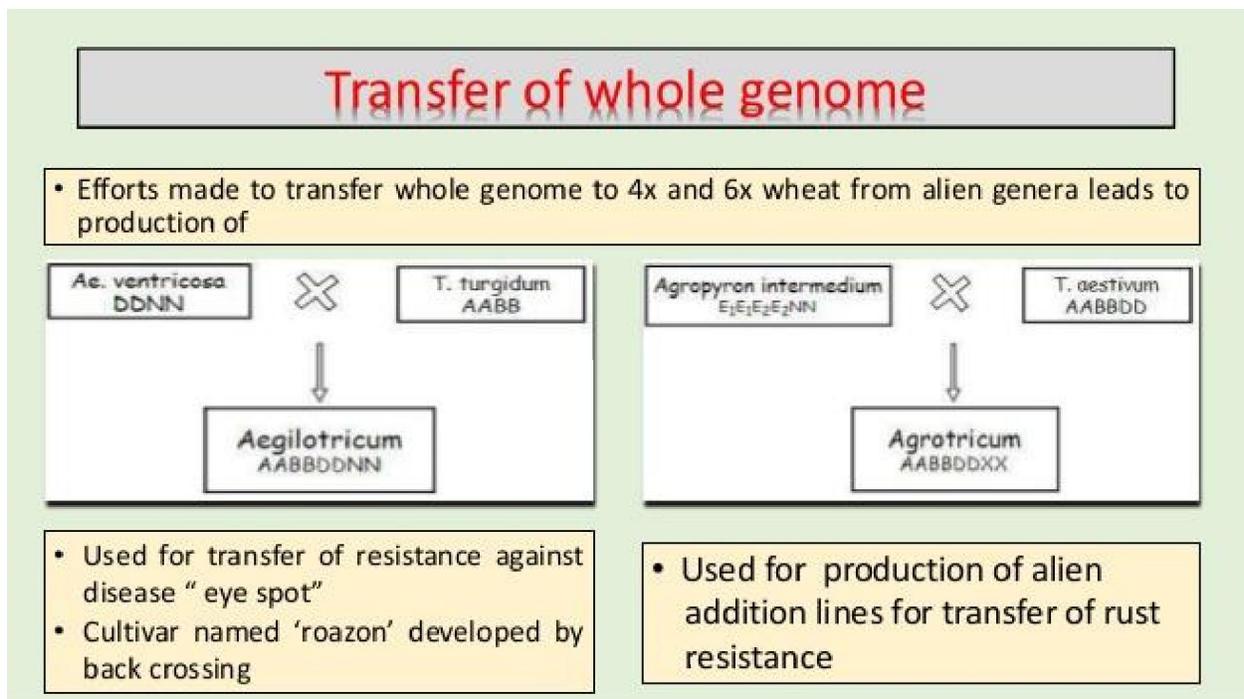
Genetic-induced gene transfers

The failure of alien chromosomes to pair with their corresponding chromosomes in the cultivated species is not always associated with structural differentiation of the chromosomes but with the genetic control of chromosome pairing. In wheat, regular bivalent pairing is controlled by the Ph gene on the long arm of chromosome 5B. The Ph gene restricts pairing to homologous chromosomes and the homoeologues of the three constituent genomes synapse and form chiasmata associations only when chromosome 5B is deleted. The presence of such a genetic system in natural allopolyploids is also responsible for minimizing pairing between alien chromosomes and their homoeologues in the crop species. The relaxation of the control mechanism leads to pairing between alien chromosomes and the corresponding wheat chromosomes. By manipulating the control mechanism to allow homoeologous chromosomes to pair have been able to induce alien chromosomes to pair with their homoeologues in wheat and isolate recombinants including genes located on the alien chromosome. Riley et al.(1968) used

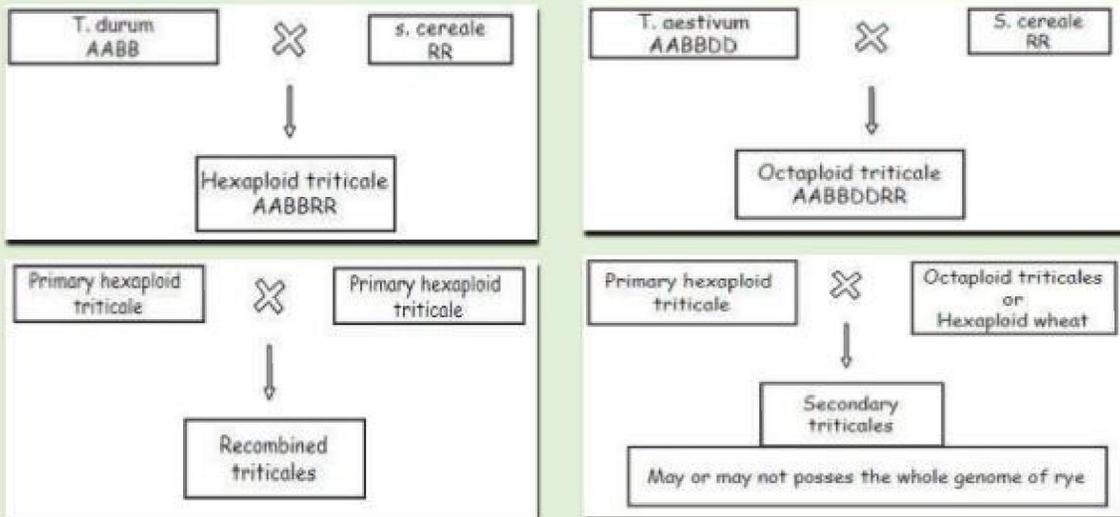
genotypes of *Aegilops speltoides*, which suppresses the activity of the Ph gene, while Sears (1973), using the appropriate aneuploid lines, deleted chromosome 5B to induce pairing between alien chromosomes and their homoeologues in wheat.

Irradiation -induced translocations:

Sears (1956) was the first to use irradiation-induced translocations to transfer resistance to brown rust from *Aegilops umbellulata* to bread wheat. The technique relies on chromosomes rejoining in novel combinations after chromosome breakage induced by the irradiation treatment. Sears (1956) irradiated spikes, just before anthesis, of plants with additions of isochromosomes of *Aegilops umbellulata* carrying a gene for resistance to brown rust, and isolated gene transfers in subsequent progeny. Irradiation of dry seeds of the disomic alien addition can also be successfully used to induce translocations (Driscoll & Jensen 1963). In gene transfers based on irradiation-induced translocations there is a deletion of part of the genome of the recipient species and a duplication arising from the alien segment introduced. The usefulness of the gene transfers will depend on the ability of the genotype to tolerate such deletions and duplications.



Triticale, a successful man made cereal

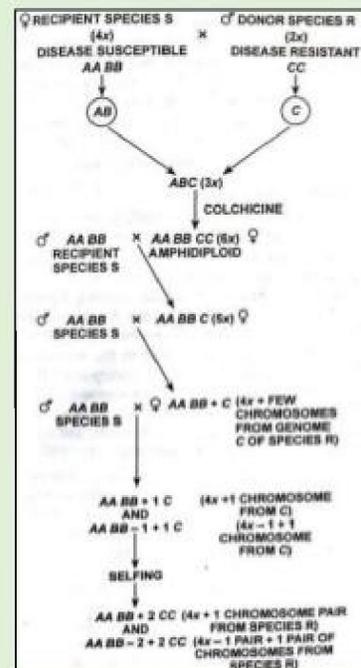


Transfer of whole chromosome

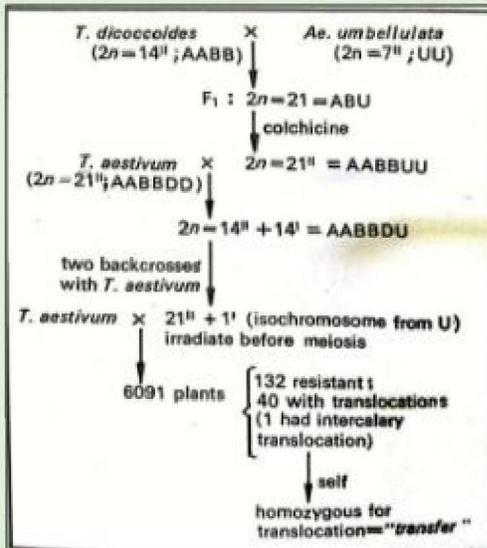
(Alien addition and substitution lines)

• Alien addition lines and alien substitution lines produced by

1. crossing of recipient species with donor
2. Produced F_1 chromosomes doubled
3. Back crossed with recipient species
4. It gives pentaploid individuals
5. They back crossed with recipient species
6. It produces alien addition monosome or substitution monosome
7. Selfing of alien addition monosome gives alien addition line
8. Selfing of alien substitution gives alien substitution line



Interchanges (using irradiation)



- Sears in 1956
- First useful transfer by irradiation
- *Ae. umbellulata* chromosome 6U to wheat chromosome arm 6BL
- Transfer of leaf rust resistance gene Lr 9
- *T. dicoccoides* used as bridging species
- Irradiation of pollen with X rays

Interchanges through recombination

- **Through manipulation of diploidizing system**
 1. Removal of 5B chromosome
 2. Suppression of 5B effect by genome of *Ae. speltoides* or *Ae. mutica*
 3. Utilizing recessive mutant of Ph1 locus on 5B
- **Without manipulation of diploidizing system**

Transfer of genome in the genus *Arachis*

Cultivated peanut is an allotetraploid ($2n = 4 \times = 40$ chromosomes) with two genome types, A and B, which are found separately in the wild species of the *Arachis* section. The A-genome species are diploids characterized by the presence of a so-called A chromosome pair, of reduced size and with a lower level of euchromatin condensation in comparison to the other chromosomes. Diploid species of the section *Arachis* with $2n = 20$ and lacking the A

chromosome pair are usually considered to share the B-type genome, although they are much more heterogeneous and may present variant forms of this B-genome. Cultivated allotetraploid peanut *Arachis hypogaea* was originated via hybridization of two diploid wild species, probably *A. duranensis* (A-genome) and *A. ipaënsis* (B-genome), followed by a rare spontaneous duplication of chromosomes. The resulting tetraploid plant would have been reproductively isolated from its wild diploid relatives. This isolation, coupled with the origin through a probably single hybridization event, leads to a limited genetic diversity of peanut, as observed in different studies using molecular markers. In contrast, wild diploid *Arachis* species are genetically more diverse providing a rich source of variation for agronomical traits, and DNA polymorphisms for genetic and genomic studies.

Although the cultivated germplasm contains a large amount of genetic variability for various traits, it lacks sources of resistance to several important pests and pathogens. Nevertheless, wild *Arachis* species with genes conferring high levels of resistance to these diseases have been identified. The development of strategies to utilize wild species has become extremely important in the genetic improvement of groundnut. utotetraploids of wild peanut species have either AAAA or BBBB genomes, so crosses with cultivated peanut (AABB genomes) are partially sterile. The method of crossing *A. hypogaea* with autotetraploids does not work well with peanut.

Crossing AABB synthetic polyploids derived from A- and B-genome diploid species with cultivated peanut is the preferred method to introgress wild species germplasm, although several generations of selection are still required to stabilize genomes and obtain highly fertile progenies. The method of choice for peanut is to first produce A-genome \times B-genome species crosses and then double the chromosome number of the sterile AB genome F₁ hybrid. Seeds obtained from the colchicine-treated cuttings will all be semifertile AABB genome amphidiploids that can then be used as a male parent in crosses with *A. hypogaea*.

Using the approach of making AABB genome synthetic polyploids, the species *A. duranensis* and *A. ipaënsis* were hybridized, the chromosome number was doubled, and the hybrid was crossed to the cultivar Fleur 11 to study genomewide segment introgression of peanut species into the cultivated genome ([Foncéka et al., 2009](#)). Marker-assisted selection was used in the BC₁F₁ and BC₂F₁ generations to select lines with donor segments and the highest percentage of the cultivated genome. Their work is leading to broadening of the genetic base of the cultivated peanut and has provided a proof of concept for introgressing genes in a systematic approach from diploid species into cultivated peanut.

An example of success with producing synthetic amphiploids are TxAg-6 and Tx Ag-7 ([Simpson et al., 1993](#)), which originated from the complex hybrid $\{A. batizocoi$ (B genome) \times [*A. cardenasii* (A genome) \times *A. diogoi* (A genome)] $\}^{4x}$. TxAG-6 has very good nematode resistance, but also significant linkage drag, which resulted in low yields and poor pod and seed quality. The RFLP markers linked to the nematode gene conferring resistan

4 cultivated species of Arachis are

- **Diploid species ($2n=2x=20$)**
 - A villosulicarpa*
 - A repens* (fodder & cover crop)
- **Tetraploid species ($2n=4x=40$)**
 - A glabrata* (fodder & cover crop)
 - A hypogaea* (oil seed) - Segmental amphidiploid
- 33 species named
 - A. monticola*** is only a wild Arachis taxon which can be crossed with *A hypogaea* to produce fertile progeny

Techniques for gene Transfer

- Transfer of whole genome
- Transfer of individual whole chromosome
- Substitution of alien chromosome arms
- Interchanges
 - a) Through irradiation
 - b) Through recombination
 - i. Manipulation of diploidizing system
 - ii. Without manipulation of diploidizing system

