Hybrid breeding - sunflower

Milan Jocković

ORIGIN OF SUNFLOWER

≈ 3000-4000 years B. C.

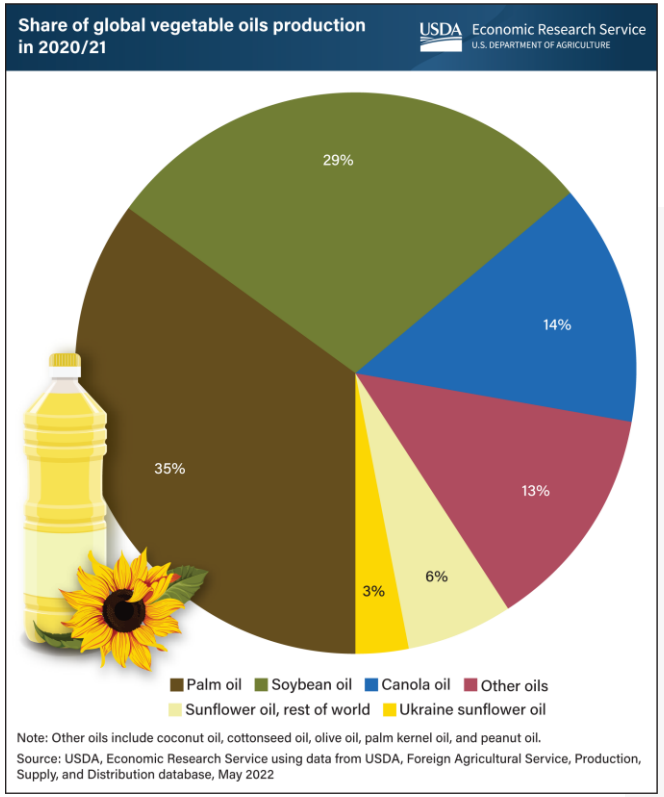
Mexico and southwest of North America

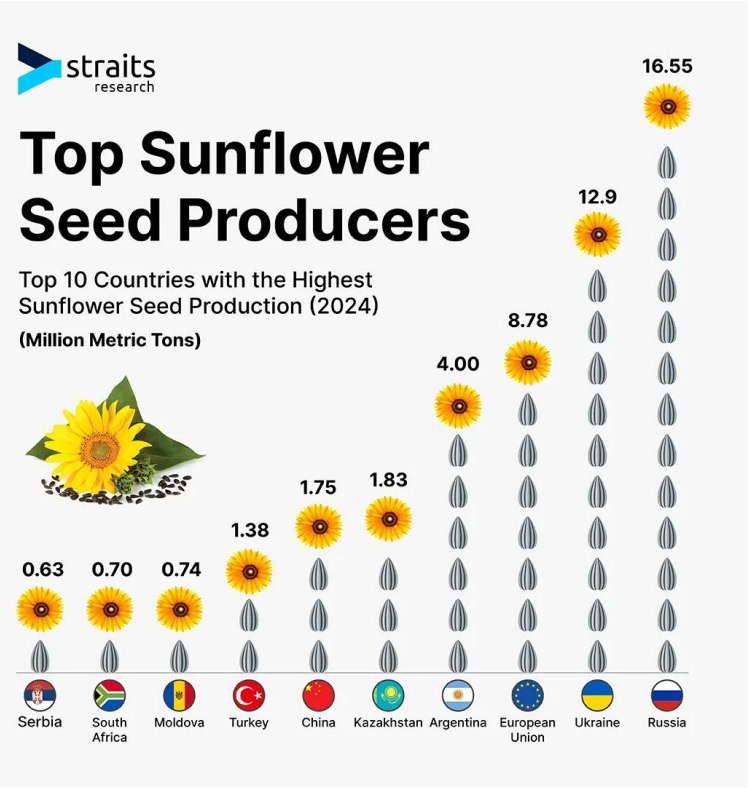
SUNFLOWER IMPORTANCE

- One of the most important source of edible oil, rich in vitamine E, cholesterol-free

- High protein meal for livestock

- Pollinator friendly





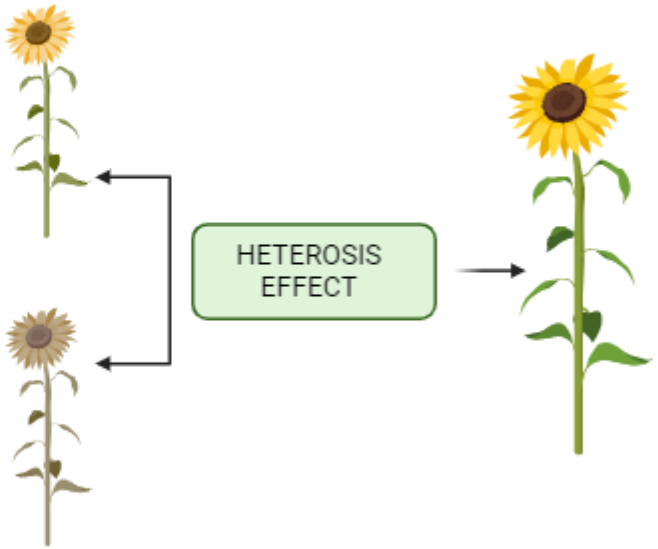
Why hybrids?

Productivity:
Seed yield
Oil yield

Enhanced resistance and quality

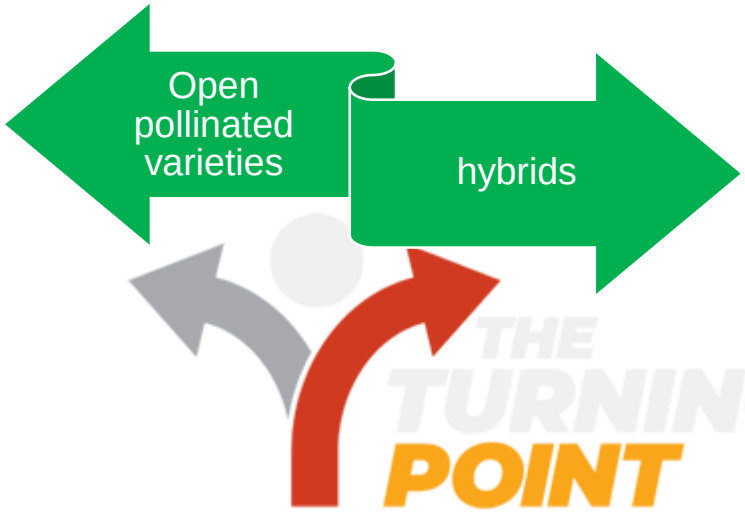
Uniformity
Germination and emergence
Flowering
Harvest

- To breed hybrid seeds, two homozygotic, but as genetically different as possible, parental lines are cross-bred with each other.
- Because of the heterosis effect, the resulting heterozygous offspring ("hybrids") are much more productive than both parents.
- The hybrid vigour makes plants bigger, more fruitful and more resilient than their parental lines.
- However, it is maintained only for one generation. Hybrid breeding is used worldwide for many crops.



History of sunflower hybrid breeding

- 1900s – sunflower breeding
- Local landraces, open pollinated varieties (OPV)
- Peredovik



1970s

- 1969 – Leclercq discoverd CMS originating from an interspecific cross between *Helianthus petiolaris* and *H. annuus* (PET-1)
- 1970 – Kinman identified the Rf1 gene in the sunflower line T66006-2-1-B which restores cytoplasmic male sterility (CMS)

Breeding methods

- Cross breeding: Crossing different varieties of a crop to bring together diverse genetic traits.
- Utilizing Diverse Germplasm: Agricultural researchers collect and use diverse genetic material, including wild relatives and landraces, which contain valuable genes.
- Induced Mutagenesis: Using chemicals or other methods to induce mutations and increase genetic variation within a plant population
- Genetic Engineering: Creating new genetic variations through modern biotechnological methods.

Cross breeding – basic steps in hybrid breeding

- Inbred line breeding
- Experimental hybrids (testing general and specific combining abilities of inbred lines)

Initial genetic material for breeding

- Local populations
- Open pollinated varieties (Op)
- Crop wild relatives (CWR)
- Interspecies populations
- Synthetic populations
- Developed inbred lines

INBRED LINE BREEDING

- ## Creating new genetics

Breeders want to provide farmers with optimally adapted crops.
This requires combining many positive characteristics in one single variety.

Breeding aims:

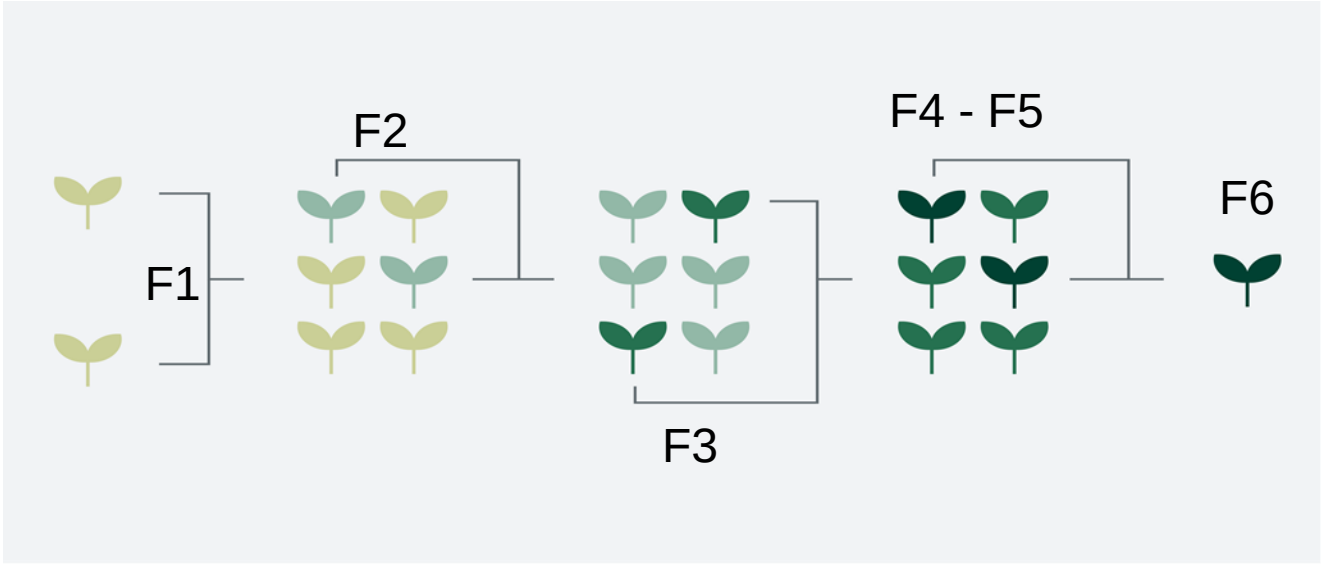
- High seed yield
- Oil content
- Protein content
- Vegetation period
- Resistance to pests
- Herbicide tolerance
- Architecture
- Tolerance to abiotic an biotic factors
- Tolerance to lodging
- Tolerance to drought

Parental plants bearing the desired characteristics are crossed with each other.

Selection (screening) – Pedigree method

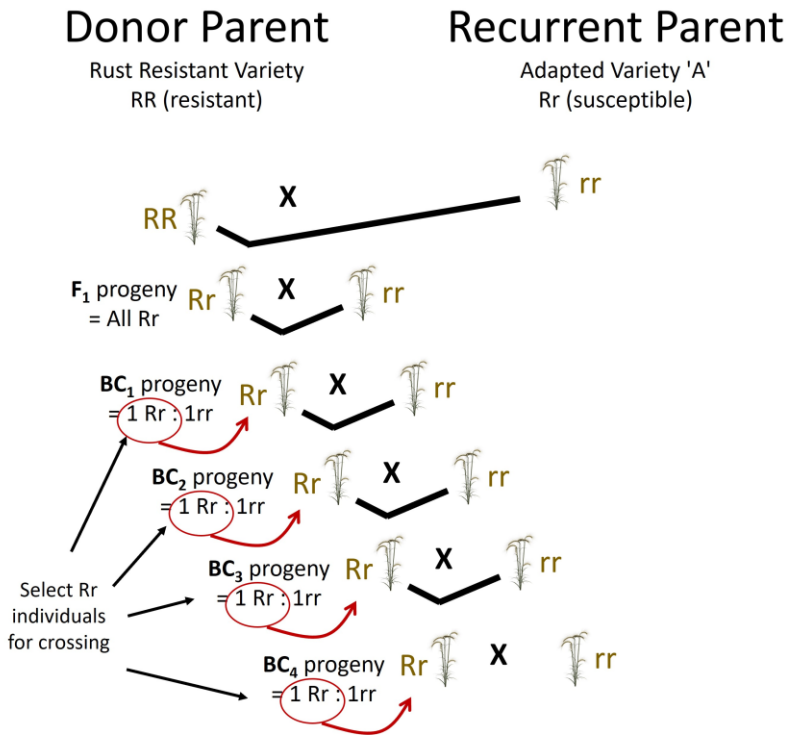
Pedigree method

- Selected parent plants with desirable traits are crossed to produce F1 seeds.
- F1 seeds are planted to produce F1 plants, which are then allowed to self-pollinate and produce F2 seeds.
- F2 seeds are harvested and space-planted to allow for the selection of individual plants with superior traits.
- It should be at least 100 plants in F2! **Detailed screening and record of each plant and selection of individual plants!**
  Field screening, laboratory screening (oil content, protein content, resistance to pests....)
- Seeds from each selected F2 plant are grown as a progeny row in the F3 generation.
  Individual plants are then selected from these rows.
- This selection and recording process continues through subsequent generations (F3-F5).
- Selection continues until the progeny rows exhibit no genetic variation, indicating that the lines are homozygous.
  Usually F6



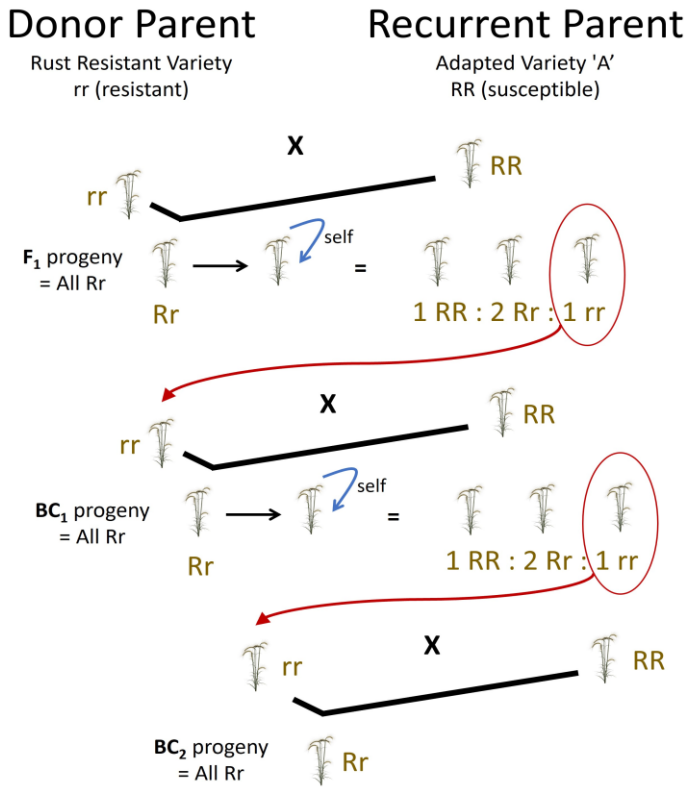
- Enhacement breeding
- Improvement of commercial inbred lines

Dominant Trait Backcrossing

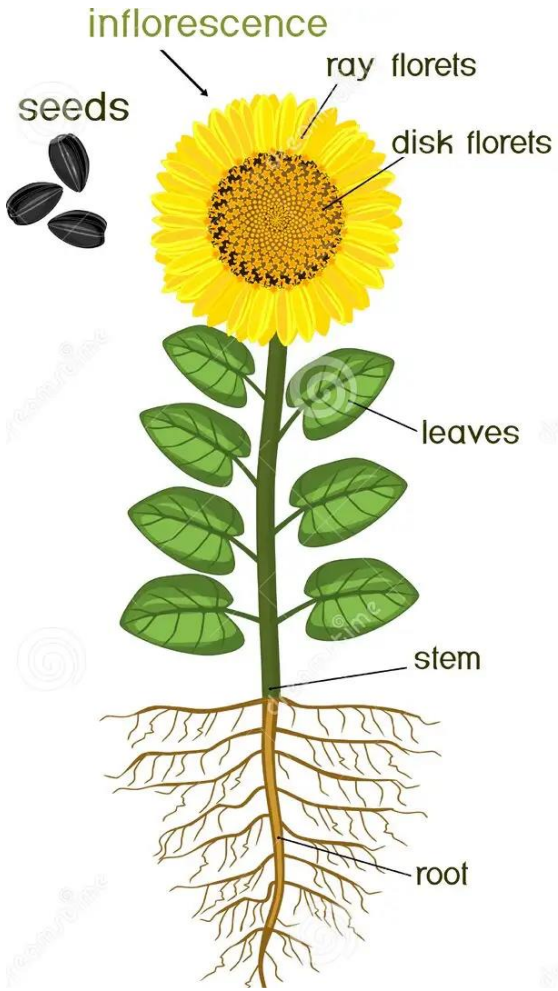


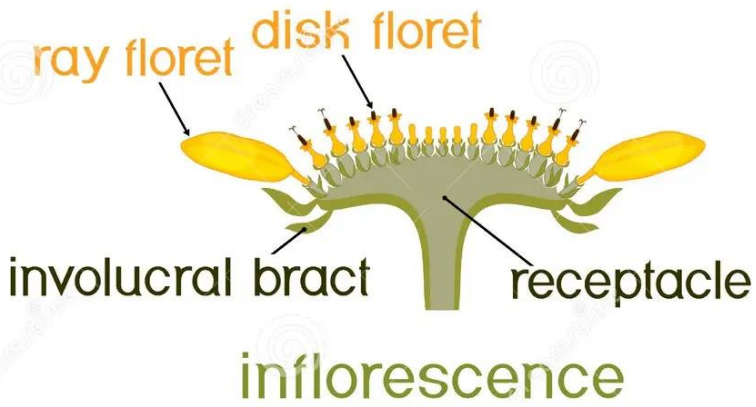
Selfing and selection for RR

Recessive Trait Backcrossing



Process continues... BC6F2


inflorescence
ray florets
seeds
disk florets
leaves
stem
root


ray floret
disk floret
involucral bract
receptacle
inflorescence


stigma
style
anther
corolla
sepal
ovary
bract
disk floret

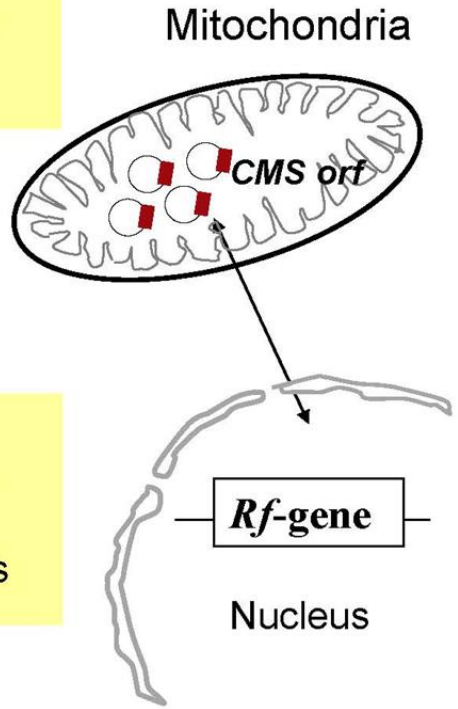

corolla
sepal
ovary
bract
ray floret

1000 - 2000

Cytoplasmic male sterility (CMS)

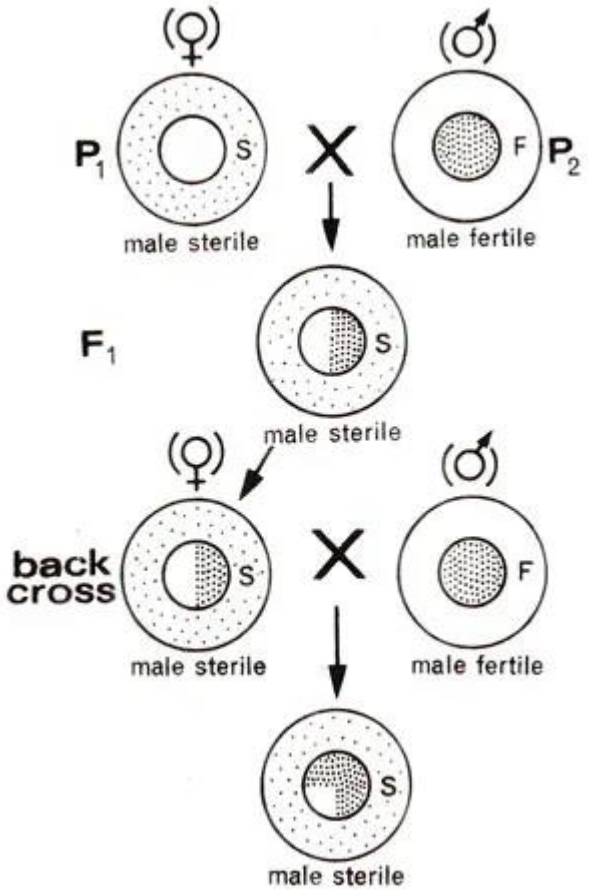
- no functional or shed pollen
- maternal inherited trait
- based on changes in the mitochondrial DNA

- restoration of fertility by dominant nuclear gene/s
- interactions between mitochondria and nucleus



Reddemann and Horn 2018. Int. J. Mol. Sci.

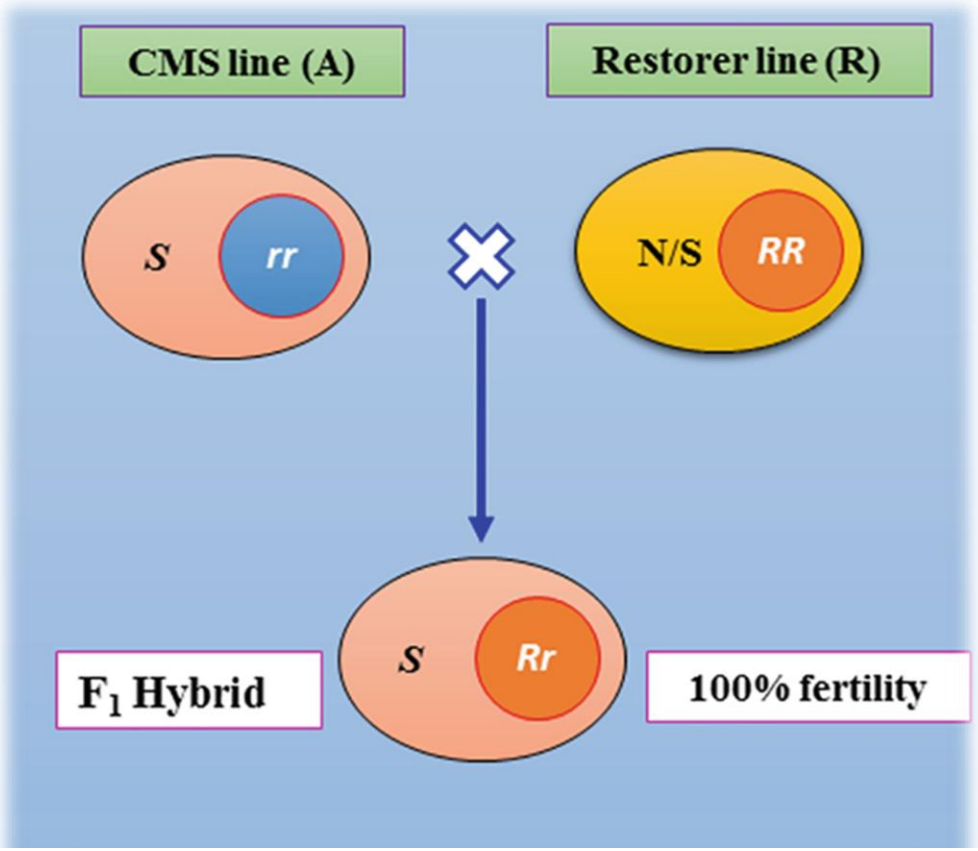
CMS conversion of maintainer (B-line)



6 back crosses

A-line

Obtaining sunflower hybrid



Testing combining abilities of newly developed lines

- General combining abilities (GCA)
- Measures the additive gene effects of a parent across all crosses. A high GCA indicates a line is a good general combiner.

- Specific combining abilities (SCA)
- Measures the non-additive gene effects for a specific cross combination. A high SCA suggests a specific cross will result in a superior hybrid.

- Testing designs

- Diallel
- Topcross
- Polycross
- Line x Tester
- Modern techniques (genomic prediction)

B-line testing

- Selected B lines (CMS form) are crossed with restorer lines (usually 3) as testers

Production of experimental hybrids

R-line testing

- Selected restorer lines are crossed with choosen B-lines (CMS form) as testers

Experimental design for testing newly developed (experimental) hybrids

- Completely randomized design (CRD) - Used when the trial site is uniform
- Randomized block design (RBD) - A highly common choice where plots are grouped into blocks with similar environmental conditions
- Latin Squares (LS) - Design used to account for both row and column variation in a field
- Split-Plot design - Ideal for studying interactions between different levels of treatments
- Augmented design - A statistical experimental design for comparing new treatments with control treatments using a large number of unreplicated entries

- Statistical analysis

- Analysis of variance (Anova) - Use ANOVA to determine significant differences between treatments
- Principal Component Analysis (PCA) - Employ PCA to analyze the relationships between different traits and provide a comprehensive understanding of the variability within the inbred lines.
- Genotype and Genotype by Environment Interaction (GGE) - Use GGE biplots for multi-environment trial (MET) data analysis to identify superior inbred lines and select those that are stable and widely adapted across different environments
- Aditive Main Effect and Multiplicative Interaction (AMMI) - a statistical technique used primarily in plant breeding to analyze genotype-by-environment (G×E) interactions in multi-location trials

Modern breeding techniques

•**Next-Generation Sequencing (NGS):**
•Allows breeders to sequence and analyze an entire plant's genome, identifying genes linked to important traits.

•**CRISPR-Cas9 Genome Editing:**
•A precise tool to modify specific genes, enabling the addition, deletion, or alteration of traits for improved yield, resilience, and pest resistance.

•**Molecular Markers: Marker assisted selection (MAS)**
•DNA markers like Single Nucleotide Polymorphisms (SNPs) help identify genomic regions associated with desirable traits.

•**Genomic Selection (GS):**
•Uses molecular markers across the entire genome to predict the breeding value of plants and select parents for breeding programs.

•**High-Throughput Phenotyping (HTP):**
•Involves automated platforms and technologies that quickly and accurately measure the physical characteristics (phenotypes) of plants, bridging the gap between genotype and phenotype.

•**Speed Breeding:**
•Uses artificial lighting and controlled environments to accelerate the growth cycle of plants, allowing for more generations to be grown in a shorter time. (green house, chambers)


THANK YOU
FOR YOUR
ATTENTION