

Lecture: - 15

Heterosis

- The term heterosis was first used by Shull in 1914.
- **Heterosis**:- It may be defined as the superiority of an F₁ hybrid over both its parents in terms of yield or some other character. Generally, heterosis is manifested as an increase in vigour, size, growth rate, yield or some other characteristic
- **Average heterosis or relative heterosis**:-The superiority of F₁ is estimated over the average of the two parents, or the mid - parent. If the hybrid is superior to the mid -parent, it is regarded as heterosis.
- **Standard/ Commercial Heterosis**:- When heterosis is estimated over standard commercial hybrid it is called as standard heterosis.
- **Heterobeltiosis**:-When heterosis is estimated over the superior parent; such an estimate is sometimes referred to as heterobeltiosis.

15.1 Manifestations of Heterosis:-

- Heterosis is the superiority of a hybrid over its parents. This superiority may be in yield, quality, disease and insect resistance, adaptability, general size or the size of specific parts, growth rate, enzyme activity, etc. These various manifestations of heterosis may be summarised as follows.
- **Increased yield**:- Heterosis is generally expressed as an increase in the yield of hybrids. The yield may be measured in terms of grain, fruit, seed, leaf, tubers or the whole plant.
- **Increased Reproductive ability**:- The hybrids exhibiting heterosis show an increase in fertility or reproductive ability. This is often expressed as higher yield of seeds or fruits or other propagules, e.g., tuber in potato (*S. tuberosum*), stem in sugarcane (*S. officinarum*), etc.
- **Increase in Size and General Vigour**: The hybrids are generally more vigorous, i.e., healthier and faster growing and larger in size than their parents. Some examples of increased size are increases in fruit size in tomato, head size in cabbage, cob size in maize, head size in jowar, etc.

- **Better Quality:**-In many cases, hybrids show improved quality. This may or may not be accompanied by higher yields. For example, many hybrids in onion show better keeping quality, but not yield, than open-pollinated varieties.
- **Earlier Flowering and Maturity:**- In many cases, hybrids are earlier in flowering and maturity than the parents. Earliness is highly desirable in many situations, particularly in vegetables. Many tomato hybrids are earlier than their parents.
- **Greater Resistance to Diseases and Pests:**- Some hybrids are known to exhibit a greater resistance to insects or diseases than their parents.
- **Greater Adaptability:**- Hybrids are generally more adapted to environmental changes than inbreds. In general, the variance of hybrids is significantly smaller than that of inbreds. This shows that hybrids are more adapted to environmental variations than are inbreds. In fact, it is one of the physiological explanations offered for heterosis.
- **Faster Growth Rate:**-In some cases, hybrids show a faster growth rate than their parents. But the total plant size of the hybrids may be comparable to that of parents. In such cases, a faster growth rate is not associated with a larger size.
- **Increase in the Number of Plant Part:**- In some cases, there is an increase in the number of nodes, leaves and other plant parts, but the total plant size may not be larger. Such hybrids are known in beans (*P. vulgaris*) and some other crops.

15.2 Prediction of the Performance of Double Cross Hybrids:

- In a double cross hybrid, four inbred parents are involved. Theoretically, the potential of the double cross will be the function of the inbreeding value of these four parental inbreds. Therefore, based on the procedure of testing of the breeding value of inbreds, the performance of a double cross hybrid can be predicted through any of the four methods indicated by Jenkins (1934). Starting with the simplest procedure these methods are:
- Top-cross testing (one cross per inbred) to know the breeding value to each of the four inbreds (total 4 top-crosses per double cross).
- Mean of the four non-parental single crosses involved in (AXB) X (CXD) double cross, viz., (AXC), (AXD), (BXC) and (BXD) (total 4 non-parental single crosses per double cross).

- Average yield performance of all possible six crosses $[n(n-1)/2]$, namely AXB, AXC, AXD, BXC, BXD and CXD (total six crosses per double cross).
- Average progeny-performance of each inbred can be determined by the mean performance of each inbred in all possible single crosses where it occurs ($n-1$ crosses per inbred). For instance, the mean performance of AXB, AXC and AXD will determine the average breeding value of the inbred A. Similarly, the mean of AXB, BXC and BXD will indicate the potential of the inbred B and so on (total 12 crosses per double cross).

15.3 Genetical Basis of Heterosis:- The genetical basis of heterosis follows two hypotheses:

A. Dominance hypothesis of heterosis. B. Over dominance hypothesis of heterosis.

Dominance hypothesis:-

- The hypothesis, first proposed in 1908 by Davenport and later in 1910 supported independently by Bruce and by Keeble and Pellew.
- Heterosis due to superiority of dominant alleles over recessive alleles. heterosis is directly proportional to the number of dominant genes contributed by each parent.
- **AABBccdd x aabbCCDDAaBbCcDd** hybrid with 4 dominant genes



- Heterosis is not due to heterozygosity *per se*.
- Superiority of dominant alleles over recessive alleles.
- Heterosis is due to masking of deleterious recessive alleles by dominant alleles.
- Heterosis is directly proportional to the number of dominant genes contributed by each parent.

Genetic Basis of Heterosis dominance hypothesis:-

Explanations:-

- Dominance of linked gene hypothesis (Jones, 1917) .
- Smooth and symmetrical as yield is governed by polygene's (Collins, 1921) .

Objections:-

- True breeding homozygous individuals for all dominant genes in F₂.
- F₂ Curve should be skewed towards dominant gene.

Over dominance hypothesis:-

- This hypothesis was proposed in 1908 by Shull and East, independently.
- It is assumed here that there is a physiological stimulus to development that increases with the diversity of the uniting gametes. In other words, using Mendelian terms, it means that there are loci at which the heterozygote is superior to either homozygote and that vigour increases in proportion to the amount of heterozygosity.
- Various workers have given different names to this hypothesis such as 'single-gene heterosis', 'cumulative action of divergen alleles' and 'stimulations of divergent alleles'. Fisher (1930) called it superdominance.

Genetic Basis of Over-dominance Hypothesis:-

- Heterozygosity per se is the cause of Heterosis
- Heterozygote is superior to better parent.
- Increase in vigour is directly proportional to diversity of combining alleles.
- Superiority of heterozygote may arise due:-
 - a) Production of superior hybrid substance in heterozygote
 - b) Cumulative action of divergent alleles
- More divergent alleles will exhibit more heterosis than less divergent ones.

Evidence:-

- They are not many clear cut cases where the heterozygote is superior to the two homozygote ; in fact over dominance has not been demonstrated unequivocally for any polygenic trait: in case in maize gene *ma* affects maturity. The heterozygote's *Ma ma* is more vigorous and later in anthesis and maturity than the homozygote's *Ma Ma*.

Objections:-

- Evidences of single locus heterosis may not hold good for quantitative characters like yield.
- Quantitative estimation of over dominance is difficult due to linkage basis.

Epistasis Hypothesis:-

- In 1952, Gowen had suggested that influence of one locus on the expression of another may be involved in heterosis. Subsequently, considerable data has accumulated to implicate epistasis as a cause of heterosis. For example, a majority of heterotic crosses show significant epistasis. But all heterotic crosses do not show epistasis, and all crosses that show epistasis are not heterotic.
- In many cases, the effects of a single homozygous successive allele is epistatic to almost the whole genetic makeup of an inbred. When the effects of such an allele are masked by its dominant allele, the effects on heterosis are usually dramatic (Stuber 1994).

INBREEDING DEPRESSION

Inbreeding Depression: - It may be defined as the reduction or loss in vigour and fertility as a result of inbreeding.

$$\text{Inbreeding depression} = \frac{F1-F2}{F1} \times 100$$

- The chief effect of inbreeding is an increase in homozygosity in the progeny, which is proportionate to the degree of inbreeding.
- The degree of inbreeding of an individual is expressed as inbreeding coefficient (F).
- The degree inbreeding is proportional to degree of homozygosity.
- East (1908) and Shull (1909) independently showed the effect of inbreeding depression while working in maize. Subsequently scientists reported inbreeding depression in other crop plants.

Effects of Inbreeding depression:-

- Inbreeding is accompanied with a reduction in vigour and reproductive capacity i.e. fertility.
- **Appearance of Lethal and Sublethal Alleles** : IB results in appearance of lethal; sublethal and subvital characters. Eg : Chlorophyll deficiencies, rootless seedlings, flower deformities.
- **Reduction in vigour** : General reduction in vigour size of various plant parts.
- **Reduction in Reproductive ability** : Reproductive ability of population decreases rapidly. Many lines reproduce purely that they can not be maintained.
- **Separation of the population into distinct lines** : population rapidly separates into distinct lines i.e. due to increase in homozygosity. This leads to random fixation of alleles in different lines. Therefore lines differ in genotype and phenotype. It leads to increase in the variance of the population.
- **Increase in homozygosity** : Each lines becomes homozygous. Therefore, variation within a line decreases rapidly. After 7-8 generations of selfing the line becomes more than 99% homozygous. These are the inbreds. These have to be maintained by selfing.
- **Reduction in yield** :IB leads to loss in yield. The inbreds that survive and maintained have much less yield than the open pollinated variety from which they have been developed.

Degrees of inbreeding depression:

Inbreeding depression may range from very high to very low or it may even be absent.

The ID is grouped into 4 categories:-

- **High inbreeding depression** : Eg : alfalfa and carrot show very high ID. A large proportion of plants produced by selfing show lethal characteristics and do not survive.
- **Moderate inbreeding depression** : Eg : Maize, Jowar and Bajra etc. show moderate ID. Many lethal and sublethal types appear in the selfed progeny, but a substantial proportion of the population can be maintained under self-pollination.
- **Low inbreeding depression** : Eg : Onion, many Cucurbits, Rye and Sunflower etc. show a small degree of ID. A small proportion of the plants show lethal or subvital characteristics. The loss in vigour and fertility is small ; rarely a line cannot be maintained due to poor fertility.
- **Lack of inbreeding depression** : The self-pollinated species do not show ID, although they do show heterosis. It is because these species reproduce by self- fertilization and as a result, have developed homozygous balance.

Procedure for development of inbred lines and their evaluation:

- Development of inbred lines: Inbred lines are developed by continues self fertilization of a cross-pollinated species. Inbreeding of an OPV leads to many deficiencies like loss of vigour, reduction plant height, plants become susceptible to lodging, insects and pests and many other undesirable characters appear.
- After each selfing desirable plants are selected and self pollinated or sib pollinated. Usually it takes 6-7 generations to attain near homozygosity. An inbred line can be maintained by selfing or sibbing. The purpose of inbreeding is to fix the desirable characters in homozygous condition in order to maintain them without any genetic change.
- The original selfed plants is generally referred as So plant and the first selfed progeny as S1 second selfed progeny as S2 as so on. The technique of inbreeding requires careful attention to prevent natural crossing.

Evaluation of inbred lines: After an inbred line is developed, it is crossed with other inbreds and its productiveness in single and double cross combination is evaluated. The ability of an inbred to transmit desirable performance to its hybrid progenies is referred as its combining ability.

- **GCA :** The average performance of an inbred line in a series of crosses with other inbred lines is known as GCA.
- **SCA :** The excessive performance of a cross over and above the expected performance based on GCA of the parents is known as specific combining ability

Thus GCA is the characteristic of parents and SCA is characteristic of crosses or hybrids. The inbreds are evaluated in following way.

Phenotypic evaluation: It is based on phenotypic performance of inbreds themselves. It is effective for characters, which are highly heritable i.e. high GCA. Poorly performing inbreds are rejected. The performance of inbreds is tested in replicated yield trials and the inbreds showing poor performance are discarded.

Top Cross test: The inbreds, which are selected on phenotypic evaluation, are crossed to a tester with wide genetic eg. an OPV, a synthetic variety or a double cross.

- A simple way of producing top cross seed in maize is to plant alternate rows of the tester and the inbred line and the inbred line has to be detasselled.
- The seed from the inbreds is harvested and it represents the top cross seed. The performance of top cross progeny is evaluated in replicated yield trials preferably over locations and years.
- Based on the top cross test about 50% of the inbred are eliminated. This reduces the number of inbreds to manageable size for next step. Top cross performance provides the reliable estimate of GCA.

Single cross evaluation: Outstanding single cross combinations can be identified only by testing the performance of single cross. The remaining inbred lines after top cross test are generally crossed in diallel or line x tester mating design to test for SCA.

- A single cross plants are completely heterozygous and homogenous and they are uniform. A superior single cross regains the vigour and productivity that was lost during inbreeding and can be more vigorous and productive than the original open pollinated variety.
- The performance of a single cross is evaluated in replicated yield trial over years and location and the outstanding single cross identified and may be released as a hybrid where production of single cross seed is commercially feasible. In case of maize the performance of single cross is used to predict the double cross performance.
 - ✓ Number of single crosses with reciprocals = $n(n-1)$,
 - ✓ Number of single crosses without reciprocals = $n(n-1)/2$

PRE BREEDING AND WIDE HYBRIDIZATION

Pre breeding: The process of initial introgression of traits from an undomesticated source or agronomically inferior source to a domesticated or adopted genotype is called as prebreeding.

- Modern cultivars i.e. high yielding varieties lead to development of uniformity which has reduces crop genetic diversity. So it is high time to diversity the genepool.
- The desired gene may be present in unwanted genepool but the transfer of gene will lead to linkage drag. Hence, the exotic germplasm often cannot be used directly in cultivar development.
- Instead the material are gradually introduced into cultivar development programme through crossing and selection of intermediates with new traits while maintaining a great amount of adopted traits.
- The traditional techniques used are hybridisation followed by back crossing or cyclical population improvement techniques. E.g. :- β carotene in tomato is obtained from *Lycopersian hirsutum*.

Wide hybridization: Hybridization between individuals from different species belonging to the same genus or two different genera, is termed as distant hybridization or wide hybridization, and such crosses are known as distant crosses or wide crosses.

- Distant hybridization consists of two types of hybridization:
- **Interspecific hybridization :-** When individuals from two distinct species of the same genera are crossed, it is known as inter specific hybridization. Eg. *Oryza sativa* x *O. perennis*
- **Intergeneric hybridization:-** When individuals being crossed belong to two different genera, it is referred to as intergeneric hybridization. E.g. Wheat x rye.
- **Objective:-**To transfer some desirable character from wild relatives that are not available in cultivated varieties. E.g. Many disease resistance and, insect resistance genes

Wide adaptability: (i.e. drought-resistance, cold tolerance etc.), Quality improvement (E.g. Cotton (fibre) Tobacco (leaf), Yield improvement (E.g. Oats, Tobacco, Maize, S. cane), Other characters (E.g. CMS, Earliness, dwarfness morphological characters)

- Exploitation of luxuriance (heterosis) in vegetatively propagated / ornamental crops. Prolonged vegetative period, Prolonged blooming period
- Creation of Novel genotypes : New species or F1 hybrids hitherto non-existent in nature.

Barriers to the production of distant hybrids:-

❖ Failure of zygote formation / cross incompatibility:-

- Inability of the functional pollens of one species or genera to effect fertilization of the female gametes of another species or genera is referred to as cross incompatibility. It may be due to:
 - failure of fertilization, because the pollen may not germinate.
 - Pollen tube is unable to reach to embryo sac and hence sperms are not available for fertilization
 - The style of the female parent may be longer than the usual length of the pollen tube growth therefore the pollen does not reach the embryo sac.
 - Pollen tube may burst in the style of another species Eg. *Datura*
 - Pollen tubes of polyploidy species are usually thicker than those of diploid species.

❖ Failure of zygote Development / Hybrid inviability:-

- The inability of a hybrid zygote to grow into a normal embryo under the usual conditions of development is referred to as hybrid inviability. This may be due to:
 - **Lethal genes:-** some species carry a lethal gene, which causes death of the interspecific hybrid zygote during early embryonic development. E.g. *Aegilops umbellulata* carries a lethal gene with 3 alleles against diploid wheats.
 - **Genetic Disharmony between the two parental genomes:-**The genetic imbalance between the two parental species may cause the death of embryos. E.g. Cotton - *G.gossypoides* x other *G. sps.* Brassica – *B.napus* x *B.oleracea*.
 - **Incompatible cytoplasm:-**Embryo development may be blocked by an incompatibility between cytoplasm of the species used as female and the genome of the species used as

male. Such an interaction, more generally, leads to hybrid weakness and male sterility in the hybrids or may sometimes leads to failure of embryo developments.

- **Endosperm Abortion:**-Seeds from a large number of distant crosses are not fully developed and are Shrunken due to poorly developed endosperm. Such seeds show poor germination, and may often fail to germinate. When the endosperm development is poor or is blocked, the condition is generally known as endosperm abortion.
- **Chromosome elimination:**-In some cases of distant hybridization, chromosomes are gradually eliminated from the zygote. This generally does not prevent embryo development, but the resulting embryo and the F1 plants obtained from such crosses are not true interspecific hybrids since they do not have the two parental genomes in full. Generally. Chromosomes from one are successively eliminated due to mitotic irregularities. e.g.

Hordeum bulbosum x *H. Vulgare*

Hordeum bulbosum x *Triticum aestivum*

Triticum aestivum x *Zea mays*

❖ **Failure of Hybrid seedling development / Hybrid sterility:**

- Some distant hybrids die during seedling development or even after initiation of flowering. The mechanisms involved in the failure of seedling development most likely involve complementary lethal genes. e.g. 1. In cotton-certain interspecific hybrids appear normal, but die in various stages of seedling growth; some plants die at flowering. Interspecific and intergeneric F1 hybrids of wheat show both chlorosis and necrosis.
- **Hybrid sterility:** Hybrid sterility refers to the inability of a hybrid to produce viable off spring. The main cause of hybrid sterility is lack of structural homology between the chromosomes of two species.

Techniques for production of distant hybrids:

- **Choice of parents:** Genetic differences exist among parents in a species for cross compatibility. More compatible parents should be selected for use in wide crosses.
- Pollinating sufficiently large no. of flowers.
- **Reciprocal crosses:** it is better to attempt reciprocal crosses when distant crosses are not successful. e.g.: *Phaseolus aureus* and *p.mungo* are crossable only when *P. aureus* in used as

female and *P. mungo* as male. Determine the barrier and then take measures to overcome it: Longer style sps - cut the style. Use more than one strain of each sps for lethal genes Auto polyploidy (*B. oleracea* x *B. compestris*). Manipulation of ploidy – when two species of a cross differ in chromosome number, it is necessary to manipulate their ploidy. Direct crossing-use higher ploidy sp as female parent. Chromosome no. of the wild species or of the interspecies hybrid (F1) may be doubled to overcome sterility of the hybrid.

- **Bridge crosses:** Some times, two species say 'A' and 'C' do not cross directly. In such case a third species say 'B' which can cross with both 'A' and 'C' is chosen as a bridge species. First 'B' is crosses with 'C' and then the amphidiploid is crossed with 'A'. Bridge crosses have been used in Tobacco and wheat. e.g. *Nicotiana repanda* can cross with *N. sylvestis* but not with *N. tabacum*, *N. sylvestris* crosses with both *N. repanda* and *N. tabacum*. For transfer of genes from *N. repanda* to *N. tabacum*, *N. sylvestis* is used as bridge species.
- **Use of pollen mixtures:** Cross incompatibility results due to unfavourable interaction between the protein of pistil and pollen which inhibits normal germination and growth of pollen tube. This problem can be overcome by using the mixture of pollen from compatible (self) and incompatible parents.
- **Manipulation of pistil:** In some cases, pollen tube is short and style is very long, due to species difference. Thus pollen tube cannot reach ovule to effect fertilization. In such situation either reciprocal cross should be made or the style should be cut to normal size before pollination. This technique is successful in maize – *Tripsacum* crosses, where maize style remains receptive even after cutting.
- **Use of growth regulation :** Some times, the pollen tube growth is so slow that the eggcell dies or the flower aborts before the male gametes reach the ovary. In such cases, growth regulators should be used to accelerate the pollen tube growth or to prolong the viability of the pistil. Use of growth regulators such as IAA; NAA; 2,4-D and GA3 etc; are promising in some wide crosses.
- **Large number of crosses :** The success of seed set is generally very low in widecrosses. Hence, large no. of crosses should be made to obtain crossed seeds.
- **Protoplast fusion :** The wide crosses can be obtained through protoplast fusion, when it is not possible to produce such crosses through sexual fusion.

- **Embryo culture** : This technique is being used widely to obtain viable interspecific or intergeneric hybrids. This is used when hybrid zygote is unable to develop. This technique has been successfully used in *Triticum*, *Hordeum*, *Phaseolus*, *Nicotiana*, *Gossypium*, *Lycopersicon*, *Trifolium*, *Cucurbita* etc.
- **Grafting** : Grafting of interspecific hybrid on to the cultivated species helps in making the cross successful.

Applications of wide hybridization in crop improvement:

- **Alien addition lines:** Carries one chromosome pair from a different species in addition to somatic chromosome complement. For E.g. Disease resistance in Wheat, oats, tobacco.
- **Alien substitution lines:** has one chromosome pair from different species in place of the chromosome pair of the recipient parent.
- **Introgression of genes:** Transfer of small chromosome segments with desirable genes. E.g.
 - A. Disease resistance: In Cotton transfer of black arm disease resistance from *G. arboreum* to *G. barbadense*.
 - B. Wider adaptation: Cold tolerance has been transferred from wild relatives to Wheat, onion, potato, tomato and grape.
 - C. Quality: Oil quality in oil palm was improved by genes from wild relatives.
 - D. Changing the mode of reproduction :
 - E. Yield
- **Development of New crop species** : E.g. Triticale

Sterility in distant hybrids:-

- Distant Hybrids show variable sterility ranging from complete fertility to complete sterility for e.g. *L. esculentum* x *L. pimpinellifolium* hybrid is completely fertile while sugarcane maize hybrid is completely sterile.
- Distant Hybrids are of two broad groups:
 - a) The first group includes those Distant hybrids that exhibit atleast some fertility so that they can be maintained by selfing, intercrossing among themselves or backcrossing to the parental species.

b) The second group consists of those hybrids that are completely sterile and have to be maintained clonally or by doubling their chromosome number.

The sterility of distant hybrids may be caused by cytogenetic, genetic or cytoplasmic factors.

Cytogenetic Basis of sterility: Most of the interspecific hybrids show reduced chromosome pairing and in extreme cases all the chromosomes may be present as univalents.

- The distribution of chromosome in such cases is irregular, and it leads to the formation of unbalanced gametes resulting in partial to complete sterility.
- Inter specific crosses also show rings and chains at metaphase-I (indicating translocations).
- Bridges and fragments at anaphase-I (indicating inversions)
- Loops at pachytene (indicating duplications or deletions). These cytological aberrations also reduce fertility. Fertility in such hybrids is improved by doubling their chromosome number, that is by producing amphidiploids from them.

Genetic Basis of sterility: Chromosome pairing in some interspecific hybrid is regular, but they show variable sterility which is due to genes. E.g. The F1 hybrid between foxtail millet, *setaria italica* and its wild relative. *S. viridis* showed normal pairing and regular formation of bivalents. But pollen and ovule sterilities were 70 and 50% respectively.

Cytoplasmic Basis of sterility: In some interspecific hybrids, sterility is produced by the cytoplasm. In such cases, the reciprocal crosses produce fertile hybrids. Clearly, in such cases sterility is produced by the cytoplasm such instances of hybrid sterility are known as *Epilobium*, *Oenothera*.

Limitations of Distant Hybridization:

- Incompatible Crosses
- F1 Sterility
- Problems in Creating New species
- Lack of Homoeology between Chromosomes of the Parental Species

- Undesirable Linkages
- Problems in the Transfer of Recessive Oligogenes and Quantitative Traits
- Lack of Flowering in F1
- Problems in using Improved varieties in Distant Hybridization
- Dormancy

Achievements:

- Wheat, Tobacco, Cotton
- Parbhani Kranthi: Derived from *A. esculentus* C.V. Pusa Sawani x *A. Manihot*
– Resistant to yellow mosaic vein virus, yield – Kharif : 110-120 q/ha,
Summer : 85-90 q/ha.
- Pusa Kranthi : Kharif 105-110 q/ha, Summer 75-80 q/ha.