

APPLICATION BIOLOGY – PLANT BREEDING

- It is the genetic improvement of the crop for better yields, disease resistant.
- Green Revolution was responsible for our country not only to meet our requirements in food production but also helped us to export it. The former mainly depended on plant breeding techniques for high yielding and disease resistant varieties in wheat, rice, maize, etc.
- Traditional or classical plant breeding involves hybridization of pure lines, artificial selection to produce plants with desirable characters of higher yield, nutrition and resistance to diseases.

MAIN STEPS IN PLANT BREEDING:

(i) Collection of variability:

- Wild relatives of the crop bear pre-existing genetic variability. Collection and preservation of all the different wild species, varieties are useful to control the exploitation of natural genes available in the population.
- **Germplasm** represents sum total of all the alleles of the genes present in a crop and its related species.
- The entire collection of plants / seeds containing all the diverse alleles for all genes in a given crop is called **germplasm collection**. The latter is required for a successful breeding programme.
- Germplasm is usually collected in the form of **seeds** from within the country or from other countries. Seeds are maintained at a low temperature. The stored seeds are grown periodically in the field to obtain fresh seeds. In case of fruit trees, the germplasm is maintained as trees grown in the field or in test tubes as shoot cultures by plant tissue culture.

(ii) Evaluation and Selection of Parents:

- The seeds of those plants are picked up for multiplication that have the desired traits. The selection is classified into two groups.

(a) Selection in self-pollinated crops

(b) Selection in cross-pollinated crops.

(a) Selection in self-pollinated crops:

- Superior homozygous genotypes are obtained by repeated self pollination. The self-pollinated progeny of homozygous plant constitutes a pure line. The plants of latter bear identical genotype. **e.g. wheat variety HUW 468.**

(b) Selection in cross-pollinated crops:

- Cross-pollinated crops are heterozygous for most of their genes thus they bear several different genotypes out of them, some genotypes are superior and many are inferior. Heterozygosity is maintained by the selection of superior genotypic plants and allowed them to perform crossbreed.

Cross or Hybridization among the selected Parents:

- Hybridization involves crossing of two or more types of plants for bringing their traits together in the progeny. The former includes a single cross (two plants) or multiple cross (more than two plants).
- Hybridization is of following types.

(i) **Intravarietal** : It is performed between two plants of a variety **e.g. Cross between two plants of *Brassica oleracea* var. *capitata* (cabbage).**

(ii) **Intervarietal (intra specific)** : It is performed between two varieties of a plant species **e.g. Cross between *Brassica oleracea* var. *capitata* (cabbage) and *Brassica oleracea* var. *botrytis* (cauliflower).**

(iii) **Interspecific** : It is performed between two species of a genera **e.g. development of rice variety ADT-37 from a cross between *Oryza japonica* and *O. indica*.**

(iv) **Intergeneric** : It is performed between two different genera **e.g. *Raphanobrassica*, *Triticale*.**

Resonate the Concept

- The process of hybridization has described in genetics.

(iv) Selection and Testing of superior Recombinations:

- The plants that have the desired character combination are selected among the progeny of the hybrids. These plants are self pollinated for several generation to obtain homozygosity so that the characters will not separate in the progeny.

(v) Testing, Release and Commercialization of New Cultivars:

- The newly selected lines are evaluated in the research field and recording their performance in ideal fertiliser, irrigation, etc. for their yield and other agronomic traits of quality, disease resistance, etc. After that the testing of the materials is performed in the farmer's fields, for at least three growing seasons at different locations in the country, the material is evaluated in comparison to the best available crop cultivar.
- **Agriculture** contributes about **33 percent of India's GDP** and gives employment of about **62 percent** of the population. After India's independence, one of the main challenges faced by the country was enough food production for the increasing population. The development of several high yielding varieties of wheat and rice in 1960 increased yields per unit area. This phase is often called the **Green Revolution**. **Dr. Borlaug** is known as '**Father of Green Revolution**' while **Dr. M.S. Swaminathan** is known as '**Father of Green Revolution in india**'.

Wheat and Rice:

- In 1960 to 2000 wheat production increased from 11 million tonnes to 75 million tonnes while rice production increased from 35 million tonnes to 89.5 million tonnes. It was due to the development of semi-dwarf varieties of wheat and rice.
- **Wheat:** Dwarfing gene **Norin -10** was noticed in Japan. It was picked up by American breeders. In 1963, **Borlaug** was able to breed high yielding triple dwarf or **Mexican wheats** which were resistant to lodging, common pathogens and pests. Two of these, **sonora-64** and **Lerma Roja-64** were brought to India and modified by gamma radiations.
- **Rice:** Dwarfing gene **dee-geo-woo-gen** was noticed in **Taiwan**. It was introduced in **Rice varieties** by **IRRI, Philippines** and developed high yielding early maturing **IR-8** and **IR-24 varieties**. Later on, semidwarf varieties 'jaya' and 'Ratna' were developed in Indian (**AIPMT Pre. - 2011**)
- **Sugarcane:** **Saccharum barberi** was originally grown in North India, but had poor sugar content and yield, However, **Saccharum officinarum** had higher sugar content and thicker stems but did not grow well in North India. These two species were crossed to have sugarcane varieties combining the desirable qualities of high sugar, high yield, thick stems and ability to grow in the sugarcane belt of North India.
- **Millets:** Hybrid bajara, jowar and maize have been developed in India. From hybrid varieties, the development of several high yielding varieties resistant to water stress were taken over.

TABLE

S.No.	Name of disease	Causal organism
1	Black rust of wheat	<i>Puccinia graminis-tritici</i>
2	Loose smut of wheat	<i>Ustilago tritici</i>
3	Late Blight of Potato	<i>Phytophthora infestans</i>
4	Bean Mosaic Disease	Common Mosaic Virus (CMV) and Yellow Mosaic Virus (YMV)
5	Bacterial Leaf Blight of Rice	<i>Xanthomonas oryzae</i>
6	Black rot of crucifers	<i>Xanthomonas campestris</i>
7	Root Knot of Brinjal and Tomato	<i>Meloidogyne incognita</i>

Test your Resonance with concept

- Heterosis is
 - (1) Appearance of spontaneous mutation
 - (2) Induction of mutation
 - (3) Mixture of two or more traits
 - (4) Superiority of hybrid over their parents
- Norin-10 gene is famous for
 - (1) Gigas effect
 - (2) Dwarfing effect
 - (3) Aromatic effect
 - (4) Early maturation effect
- Amber coloured Wheat is produced from red coloured Mexican Wheat is
 - (1) Sharbati sonora
 - (2) Pusa Lerma
 - (3) Both 1 and 2
 - (4) Tanchung Native 1
- Norman Borlaug is associated with
 - (1) White revolution
 - (2) Green revolution
 - (3) Blue revolution
 - (4) Yellow revolution
- Ability of plant cells to regenerate into complete plant is known as
 - (1) Pleuripotency
 - (2) Totipotency
 - (3) Tissue culture
 - (4) Cell cloning

Answers

1. (4) 2. (2) 3. (3) 4. (2) 5. (2)

Resonate the Concept

- Quarantine:** All introductions (either exotic or indigenous) are carefully examined for the presence of weeds, insects and disease-causing organisms. It is called **quarantine**. Only those introductions that are free from the above, are permitted to be used, and the rest are destroyed.
- Inbreeding Depression:** Mating of closely related individuals by ancestry. Inbreeding is useful in self pollinated plants. Loss of vigour and appearance of number of defective traits associated with inbreeding, is called **inbreeding depression**. The latter promotes homozygosity in recessive alleles that express their harmful effects.
- Heterosis or hybrid Vigour:** It is phenotypic superiority of the hybrid over either of its parents in one or more traits.

Methods of breeding for disease resistance:

Breeding is carried out by the conventional breeding techniques (described earlier) or by mutation breeding. The conventional method of breeding for disease resistance is that of hybridisation and selection. Its steps are essentially identical to those for breeding of any other agronomic characters such as high yield. The various sequential steps are - screening germplasm for resistance sources, hybridisation of selected parents, selection and evaluation of the hybrids and testing and release of new varieties.

Some crop varieties bred by hybridisation and selection, for disease resistance to fungi, bacteria and viral disease are released.

S.No.	Crop	Variety	Resistance to diseases
1	Wheat	Himgiri	Leaf and stripe rust, hill bunt
2	Brassica	Pusa swarnim (Karan rai)	White rust
3	Cauliflower	Pusa shubhra, Pusa Snowball K-1	Black rot and Curl blight black rot
4	Cowpea	Pusa Komal	Bacterial blight
5	Chilli	Pusa sadabahar	Chilly mosaic virus, Tobacco mosaic virus and Leaf curl

Mutation Breeding:

- Sudden and inheritable change in a character of an organism is called mutation.
- Various types of mutagens like Chemical mutagens, Physical mutagens cause mutations. The latter are called **induced mutations** firstly produced by **Muller (1927)** with the help of **X-rays on Drosophila** and by **Stadler in maize**.
- The development of improved varieties by the use of induced mutations in plant breeding is called **mutation breeding**.

Some useful achievements of mutation breeding are as follow.

- Red grained high yielding Mexican wheat varieties like **Sonora - 64** and **Lerma Rojo - 64** treated with gamma radiations and developed amber grained varieties **Sharbati Sonora** and **Pusa Lerma** respectively.
- Gamma ray** treatment of '**Pelita-1**' rice lines in indonesia has produced a high yielding variety called **alomita- 2** which is also resistant to **brown plant hopper**. '**Reimei**' is another high yielding Rice variety produced through **gamma irradiation**.
- Aruna variety of castor**.
- In mungbean, resistance to yellow mosaic virus and Powderly mildew were introduced by mutations.
- NP 836 variety of wheat**.

Resonate the Concept

Resistance to **yellow mosaic virus in bhindi (*Abelmoschus esculentus*)** was transferred from a **wild species *Abelmoschus manihotland*** and resulted in **new variety of *A. esculentus*** called **Parbhani Kranti**.

Plant Breeding for Developing Resistance to Insect Pests:

- Large scale destruction of crop plant and crop is due to two major causes-insects and pest infection. Morphological, biochemical or physiological characters are responsible for insect resistance in host crop plants. **e.g. Solid stems in wheat lead to nonpreference by the stem saw fly and smooth leaved and nectar-less cotton varieties does not attract boll worms. Low nitrogen, sugar and high aspartic acid in maize develops resistance to maize stem borers.**

S.No.	Crop	Variety	Insect Pests
1	Brassica (rapeseed)	Pusa Gaurav	Aphids
2	Flat bean	Pusa sem 2, Pusa sem 3	Jassids, aphids and fruit borer
3	Okra (Bhindi)	Pusa Sawani Pusa A-4	Shoot and fruit borer

Cultivated varieties, germplasm collection of the crops or wild relatives of the crop are the sources of resistant genes.

Plant Breeding for Improved Food Quality:

- More than 840 million people in the world do not have adequate food to meet their daily requirements. Three billion people suffer from protein, vitamins and micronutrient deficiencies or 'hidden hunger' because these people cannot afford to buy adequate vegetables, fruits, legumes, fish and meat.
- Breeding of crops with higher levels of vitamins and minerals or higher protein and healthier fats is called **Biofortification**.
- Objectives of improving nutritional quality of the plants by plant breeding are as follow.
 - (i) **Protein content and quality.**
 - (ii) **Oil content and quality.**
 - (iii) **Vitamin content.**
 - (iv) **Micronutrient and mineral content.**
- Wheat variety with high protein content **Atlas 66** has been used as a donor for improving cultivated wheat.
- **Maize hybrids** that had twice the amount of the **amino acids – lysine and tryptophan**, compared to existing maize hybrids were developed in 2000.
- **Indian Agricultural Research Institute (IARI)**, New Delhi, has also developed many vegetable crops that are rich in minerals and vitamins. For example, vitamin A enriched carrots, pumpkin, spinach, vitamin C enriched bitter gourd, Bathua, tomato, mustard, calcium and iron enriched spinach and bathua; and protein enriched beans (broad lablab, French and garden peas).

Resonate the Concept

- **Antinutritional factors:** These are biochemicals found in food articles that have an adverse effect on health, growth and development of human/animals. e.g. (1) **Red Kidney Bean and French Bean** bear toxic proteins **lectins** that is responsible for **agglutination of RBCs**. e.g. (2) **Khesari, (*Lathyrus sativus*)** contains **neurotoxin cyanoalanine** which causes **muscular weakness, tremor (lathyrism—a type of paralysis)**. (3) **Rape seed (*Brassica napus*)** contains **erucic acid and glucosinolates** that are harmful to human health. Some new varieties of **Rape seed** like **Canola and Hoya** have been developed that are free from both erucic acid and glucosinolates.

Single Cell Protein (SCP):

- Production of microbial biomass as supplementary food for humans and animals is called SCP. ***Spirulina, yeast, Chlorella* and *Fusarium graminearum*** are common SCP. The latter is rich in **high quality protein**, vitamins and minerals but poor in fat. SCP has to be processed to remove the excess of nucleic acids. SCP is also helpful to **reduce environmental pollution**.
- **250 g of a microorganism** like ***Methylophilus methylotrophus*** produce about **25 tonnes of protein per day** as compared to the **250 kg cow** that produces **200 g of protein per day**.

Tissue culture:

- It is the technique of maintaining and growing plant cells, tissues or organs on artificial medium in suitable containers in the presence of controlled environmental conditions.
- **Gottlieb haberlandt (1902)** firstly described tissue culture.
- The plant part that is cultured is called **explant**. The ability of any cell/explant to generate whole plant under sterile conditions in special nutrient media is called **cellular totipotency**.
- **Nutrient Media:** It is prepared by the addition of inorganic salts (both micro and macro elements), a carbon source (usually **sucrose**), vitamins, amino acids, **growth regulators –auxins like 2, 4–D, cytokinins such as BAP (Benzyl Amino Purine), gibberellins**. Other compounds like Casein hydrolysate, Coconut milk, malt extract, yeast extract, tomato juice, etc. Culture medium may be liquid, semi solid or solid.

- Both explant and Nutrient media are sterilized before culturing. Explants are sterilized by the use of disinfectants like **Sodium hypochlorite**. It is called **surface sterilization**. Containers, nutrient medium and instruments are thoroughly sterilised in **Autoclave (at 120° C) for 15–30 minutes** or by dry heat, alcohol filtration.

Types of Plant Tissue Culture:

- (1) **Callus Culture:** Explant undergoes cell division in culture medium and forms irregular unorganised and undifferentiated mass of actively dividing cells called **Callus**. Growth regulators **2, 4-D** and **BAP** are added in the medium that stimulates cell division in explant. The Callus is obtained within 2–3 weeks.

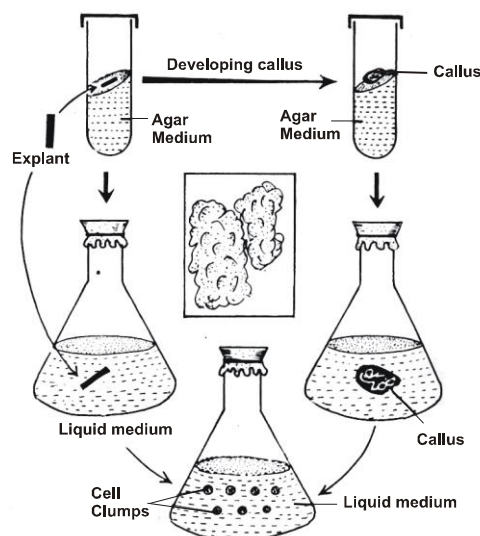


Fig :- Initiation of callus and suspension cultures.

- (2) **Suspension Culture:** In this culture single cells or small groups of cells suspended in a liquid medium containing **auxin 2, 4-D**. Now this culture is constantly agitated at 100–250 rpm (revolutions per minute). Agitation serves following three purposes.

- Aeration of culture**
- Constant mixing of medium**
- Breakage of cell aggregates into smaller groups.**

- Suspension cultures grow much faster than callus culture. Cell / tissues are regularly transferred into new culture vessels containing fresh media to avoid dryness & death of cell / tissue. This process is called **subculturing**.
- (3) **Meristem Culture:** It involves Cultivation of axillary or apical shoot meristems.
- Multiple shoot culture :** A shoot tip or bud with 1–4 leaf primordia is sterilised and introduced over culture medium with high salt content and **Benzyl Amino Purine (BAP)** after 4–6 weeks the shoot tip is given cuts or shaken to form more buds. They are excised and transferred to medium rich in auxin for induction of rooting. Plantlets are first subjected to hardening and then established in fields.

Resonate the Concept

- Virus free plants can be obtained by shoot tip culture.** Presence of high concentration of auxins and rapid rate of cell division in shoot-tip makes pathogens free to the shoot tip.

- The meristem (apical and axillary) is free of virus. Hence, one can remove the meristem and grow it in *vitro* to obtain virus-free plants. [AIPMT-2014]

The shoot apical meristem with 1–2 leaf primordia is taken and sterilised. After that it is introduced in the aseptic culture medium.

(4) Anther and Pollen Culture or Production of Haploid Plants:

- **Guha and Maheshwari (1964)** developed this technique by using **young anthers of *Datura***.
- Firstly **clorox** is used to sterilise unopened floral buds for 10–20 minutes. After that anthers are cut and introduced over culture medium. Each anther develops into number of **haploid embryoids** with in 4–6 weeks by which haploid callus and then shoots regenerate. The **young haploid plants (Androgenic haploid)** are sterile. **Colchicine treatment** changes **haploids** into **homozygous diploids** that are pure for every trait. e.g. **Jinghua-1 variety of winter wheat, Guan - 18 variety of rice** are produced by this technique.
- Androgenic haploids are pure for their characters and useful in mutation breedings.
- **Gynogenic haploids** can be formed by using **unfertilized ovules**.

Protoplast Culture and Somatic Hybridisation:

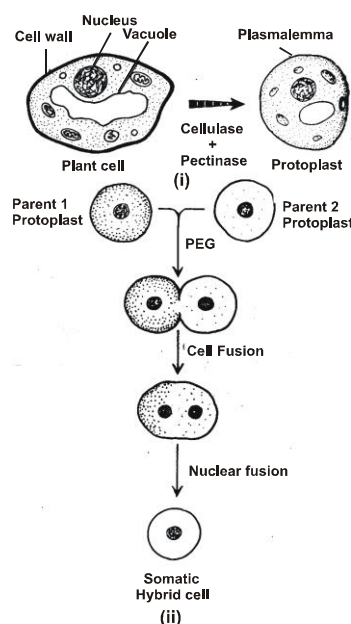


Fig:- Somatic hybridisation (i) Formation of protoplasts (ii) Protoplast fusion induced by PEG

- It involves fusion of two somatic cells of two varieties or species, The product is called somatic hybrid and process of formation of **Somatic hybrids** is called **Somatic hybridisation**.
- First of all, the cell wall of plant cells are removed by the application of **pectinase** and **cellulase** enzymes as a result protoplasts are formed.
- Now protoplasts of the two plants are introduced in the culture medium and stimulated them for the fusion either by **electric currents** or by chemicals like **PEG (Polyethylene glycol)** or **Sodium nitrate**. It forms hybrid protoplasts which may bear single fusion nucleus (synkaryon) or two unfused nuclei (Heterokaryon).
- Sometimes one of the two nuclei degenerates and the hybrid protoplasts is called **cytoplasmic hybrid** or **Cybid (heteroplasts)**.
- The first somatic hybrid was obtained by **Carlson et al (1972)** by the cross between two species of **Tobacco (*Nicotiana glauca* and *N. langsdorfi*)**. **Pomato** is somatic hybrid formed by the cross between **Potato and Tomato**. The former represents intergeneric somatic hybrids. **Bromato (Brinjal × Tomato)** is also intergeneric somatic hybrid.

Embryo Culture:

- Fragile or young or dormant embryos of developing seeds are separated and introduced in the culture medium to form seedlings and then young plants. Its applications are as follow.
 - Embryo Rescue:** It is useful technique particularly when embryo aborts at an early stage of development. The fragile embryos from fertilised ovules of interspecific crosses are taken before their abortion and culturing them to form viable hybrid seedlings **e.g. Jute (*Corchorus olitorius* × *C. capsularis*), Bean (*Phaseolus vulgaris* × *P. angustissimus*).**
 - Stored food is absent in **Orchid** seeds. Embryo culture allows seedling development in the plants.
 - Rare Plants:** The multiplication of some rare plants is performed by embryo culture **e.g. Makapuno Nut.**

Micropropagation:

- It is rapid vegetative multiplication of ornamental plants and fruit trees by using small sized explants through tissue culture technique. The propagation technique is named as micropropagation due to minute size of the propagules in the culture. Plants obtained by vegetative propagation of a single plant constitute a somaclone that have the same genotype.
- The method of micropropagation involves four steps.
 - Initiation of culture** – Introduction of explant like shoot tip on a suitable nutrient medium.
 - Shoot formation** – Formation of multiple shoots by cultured explant.
 - Rooting of shoots** – Development of roots in 'in vitro' developed shoots.
 - Transplantation** – Plantlets are treated by hardening treatment and planted in the field.

Application of Tissue Culture:

- Somaclonal Variation:** Genetic variations that occur during tissue culture are called somaclonal variations. Some of these are useful **e.g. high protein content & resistance to late blight in Potato, resistance to Tongro virus and Leaf Hopper in Rice short duration in sugarcane etc.**
- Rapid Clonal Propagation**
- Transgenic Plants**
- Resistance to Weedicides**
- Induction and selection of Mutations.**

Test your Resonance with concept

- Tissue culture medium is sterilised by

(1) Antifungal agents	(2) Keeping at 20°C
(3) Autoclaving at 120°C for 15 minutes	(4) Filtering through fine sieve
- Protoplasts of a plant cell are produced by treatment with

(1) Cellulase	(2) Pectinase	(3) PEG	(4) Both (1) and (2)
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- Explant is

(1) Plant collected after harvesting
(2) Exploited part of a plant
(3) Small part of the plant meant for tissue culture
(4) Uprooted for transplantation
- Variations appearing in tissue culture are

(1) Culture variations	(2) Auxotrophs
(3) Somaclonal variations	(4) none of these
- Vegetative propagation through cell or tissue culture is known as

(1) Macropropagation	(2) cloning	(3) Micropropagation	(4) Asexual propagation
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Answers

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|--------|--------|--------|--------|--------|
| 1. (3) | 2. (4) | 3. (3) | 4. (3) | 5. (3) |
|--------|--------|--------|--------|--------|

Resonate the Concept

1. Maximum Yield of a Cereal – Maize, that is rich in thiamine but poor in lysine and tryptophan.
2. 'Shakti', 'Rattan' and 'Protina' are lysine rich varieties of maize that have-been developed in India.
3. Oldest Cereal Crop–Barley.
4. Sporeine First commercial bioinsectide based on *Bacillus thuringiensis*.
5. Some important plant breeding centres in India.
 - (i) Central Rice Research Institute, Cuttack.
 - (ii) Central Potato Research Institute, Shimla.
 - (iii) Indian Agricultural Research Institute (IARI), New Delhi.
 - (iv) National Botanical Research Institute, Lucknow.
 - (v) Sugarcane Breeding Institute, Coimbatore.
 - (vi) Central Drug Research Institute (CDRI), Lucknow.
 - (vii) Central Food Technology Research Institute, Mysore.
 - (viii) Central Institute of Toxicology, Lucknow.
6. CDRI, Lucknow developed a Biocide from the microorganism *Bacillus sphaericus* which is highly effective formulation to control the mosquito menace.