Quadrant II – Transcript and Related Materials

Programme: Bachelor of Science (Third Year)

Subject: Botany

Course Code: BOD 101

Course Title: Plant Tissue Culture

Unit: Somatic Hybridization

Module Name: Introduction to Somatic Hybridization and its Applications

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Notes:

Introduction

In plant breeding programmes sexual hybridization is an important tool to improve cultivated crops. It involves the artificial cross-fertilization between genetically dissimilar individuals to combine several desirable traits present in different varieties into one single variety. But sexual hybridization can be performed within a plant species or very closely related species. Many desirable combinations of characters cannot be transmitted through this method of genetic manipulation. This species barriers for plant improvement which is encountered in sexual hybridization is overcome by another process which helps in genetic recombination. In this process known as somatic hybridization, crossing of plants is done through fusion of somatic cells and such hybrids are called somatic hybrids.

Somatic hybridization

Somatic hybridization involves the *in vitro* fusion of isolated protoplasts to form a hybrid cell and its subsequent development to form a hybrid plant. When a protoplast is placed in culture medium it has the capacity of cell wall regeneration, cell division and plant regeneration. Protoplasts of different species can be fused to generate a hybrid and this process is referred to as somatic hybridization or protoplast fusion and the hybrids obtained are called as somatic hybrids. In somatic hybridization, the sexual process is bypassed. Somatic hybridization helps in gene transfer between any two species irrespective of their taxonomic relationship. It overcomes the barriers of cross-incompatibility and makes incompatible crosses compatible.

Protoplasts are naked plant cells without the cell wall, but they possess plasma membrane and all other cellular components. The term protoplast was introduced by Hanstein in 1880. The first isolation of protoplasts was achieved by Klercker using mechanical method in 1892. In 1960, E.C. Cocking demonstrated that protoplasts can also be obtained by enzymatic degradation of cell walls.

Steps involved in somatic hybridization:

(i) Isolation of protoplasts

(ii) Fusion of protoplasts

(iii) Selection of hybrid cells

(iv) Culture of hybrid cells

(v) Regeneration of plants from hybrid tissues

Applications of Somatic Hybridization

Somatic hybridization is largely used for hybridization between species and genera which are not possible to cross by conventional sexual methods. In this method, various genomes of different species or genera are combined with the ability to overcome sexual incompatibility and the barrier between plants. Some important applications of somatic hybridization include:

1. Production of novel hybrids: Due to somatic hybridization, it is possible to make crosses between distant plant species and between different genera. Hybrid production is manipulated so that the desirable characteristics such as resistance to diseases, stresses, pesticides, etc. and improved protein quality, nitrogen fixation, etc. which are genetically controlled can also be transferred from one species to another. Somatic hybridization has enabled the development of unique hybrid plants which were impossible to achieve by conventional sexual hybridization. Most of the plants used in somatic hybridization belong to family Solanaceae which include *Nicotiana, Petunia, Datura, Lycopersicum* etc. Fusion of protoplasts from different species and genera has resulted in many intraspecific, interspecific (*Oryza brachyantha, O. elchngeri, O. officinalis and O. perrieri*) and intergeneric hybrids such as Pomato

(*Solanopersicon)* obtained by fusing the protoplasts of *Lycopersicum esculentum* (tomato) with *Solanum tuberosum* (potato).

2. Means of genetic recombination in asexual or sterile plants: Somatic hybridization is the only method in which two different parental genomes can be recombined among plants which cannot reproduce sexually. The protoplasts of sexually sterile i.e. haploid, triploid or aneuploid plants can be fused to produce fertile diploids and polyploids. E.g. protoplasts isolated from dihaploid *Solanum tuberosum* clones are fused with isolated protoplasts of *Solanum brevidens* to obtain hybrids of breeding value.

3. Disease resistance: Somatic hybrids obtained by fusing protoplasts from leaves of *Nicotiana nesophyla* with cell suspension culture protoplasts of an albino mutant of cultivated *Nicotiana tobacum* shows potential agricultural usefulness for disease resistance in crop plants. The somatic hybrids thus produced are resistant to Tobacco Mosaic Virus like its wild parent *Nicotiana nesophyla*. A large number of interspecific and intergeneric hybrids with disease resistance are created. Many disease resistance genes such as tobacco mosaic virus, potato virus X, club rot disease is successfully transferred from one species to another.

4. Abiotic stress tolerance: Somatic hybridization is also useful in transferring stress tolerant genes from wild relatives to high yielding varieties of crop plants. The genes responsible for tolerance to cold, frost and salt are successfully introduced through somatic hybridization. When salt tolerance in *Lycopersicum cheesmanii* and cold tolerance in *Lycopersicum hirsutum* is integrated with cultivated tomatoes, then the tomatoes develop tolerance to salt and cold. After substituting *Solanum acule* genome by *Solanum tuberosum*, an increase in frost tolerance is observed.

5. Transfer of cytoplasmic male sterility: Somatic hybridization is useful in transferring cytoplasmic traits from one species to another. Cytoplasmic male sterility (CMS) is a maternally inherited trait and the plant fails to produce viable pollen, and it is reported in a large number of plant species. Cytoplasmic male sterility is encoded in the mitochondrial genome and can arise spontaneously due to mutation in the genome (autoplasmy) or can be expressed following cytoplasmic substitutions due to nuclear-mitochondrial incompatibility (alloplasmy). Cytoplasmic male sterility phenotypes show a wide range of reproductive abnormalities, including degenerate anthers, aborted pollen, carpelloid and petaloid stamens. Somatic hybridization via protoplast fusion is a possible alternative for gene transfer from wild relatives to crops, by combining both nuclear and cytoplasmic genomes of two distantly related species, genera or even tribes.

6. Improvement in quality characters: Somatic hybrids for the production of high nicotine content in *Nicotiana tobacum* and *Nicotiana rustica*, and low erucic acid in *Brassica napus* and *Eruca sativa* are obtained.

Limitations of Somatic Hybridization

Although somatic hybridization is a novel approach in plant biotechnology, there are several limitations:

1. Somatic, hybridization does not always produce plants that give fertile seeds in all instances.
2. Regenerated plants obtained from somatic hybridization are often variable due to somaclonal variations, chromosomal elimination, organelle segregation etc.
3. Some of the somatic hybrids, particularly when produced by the fusion of taxonomically different partners, are unbalanced and not viable.
4. There is no certainty as regards the expression of any specific character in somatic hybridization. Such hybrids have many undesirable traits of wild or unrelated species.
5. Somatic hybridization between two diploids results in the formation of an amphidiploid which is not favourable. For this reason, haploid protoplasts are recommended in somatic hybridization.
6. Generally, intergeneric and intertribal hybrids exhibit high degree of sterility and cannot be backcrossed with cultivated species.