Course- B.Sc. (Honours), Part -II

Subject- Botany, Paper-IV (Group-A)

Topic- Importance of embryo culture

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The importance and application of embryo culture on the basis of available information can be broadly grouped into three categories:

1. Importance of embryo culture in relation to biological knowledge
2. Applied aspects of embryo culture
3. Other applications

1. Importance of Embryo Culture In Relation to Biological Knowledge:

In vitro embryo culture has been usefully employed to study some very fundamental problems which are difficult to undertake in embryos enclosed within seeds. Experiments with embryos separated from the seeds without interference from accessory tissues have contributed a lot of information's to our biological knowledge of plant development in the following aspects.

(i) It helps determining the factors that regulate the growth of the primodial organs of the seedling plant.

(ii) It helps to study the metabolic and biochemical aspects of dormancy and germination.

(iii) It helps in analysis of the various parameters of embryonic growth.

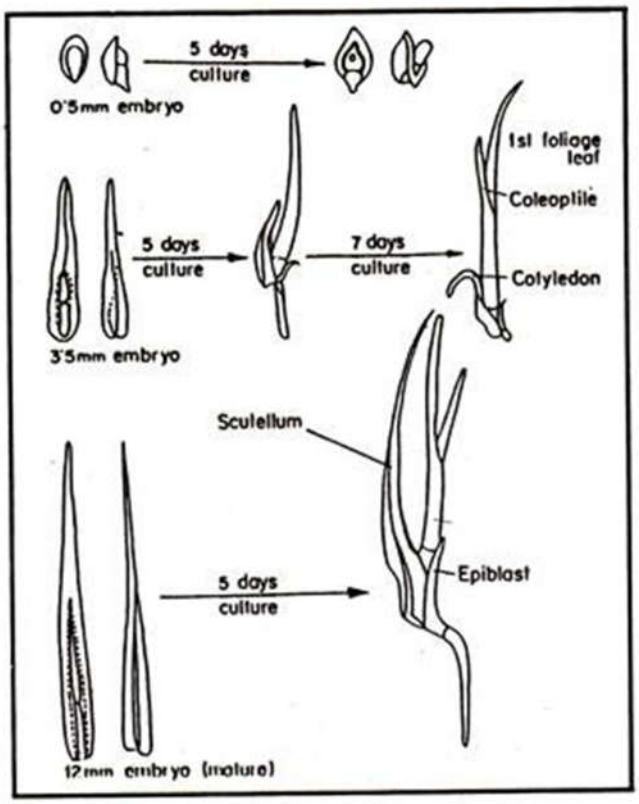
(iv) The culture of pro embryo helps to understand the control of differentiation and the nutritional requirements of progressively smaller embryos.

(v) The culture of surgically dissected embryos segments has facilitated understanding the relationship of the different parts of the embryo to its final form in culture.

(vi) An embryo undergoes a gradual transition from the dependence of the zygote to the relatively autonomy of the mature embryo. Changes in nutritional requirements of embryos at different stages can be demonstrated by embryo culture.

(vii) The role of accessory tissues can be studied by culturing isolated embryos.

In vitro microsurgical experiments with embryos of cassytha filiformis have shown that cotyledon play an important role in the development of seedling (Fig 10.5). It is evident from the data of Rangaswamy and Rangan (1971) that in C. filiformis the growth factor(s) for shoot development resides in the "radicular-halves" of the cotyledon. If the "plumular halves" of both the cotyledons are removed the seedling is well developed but if redicular halves are removed the plumule does not grow.

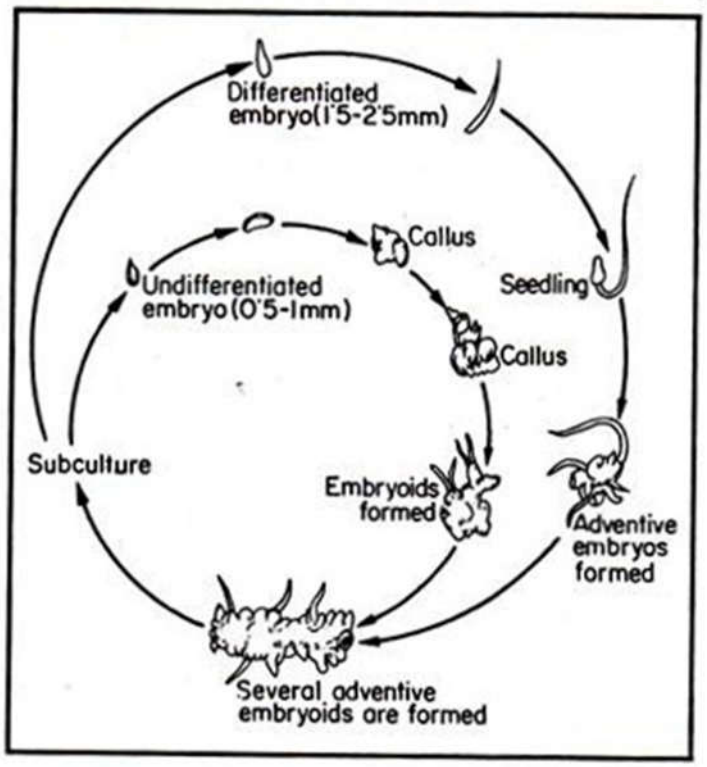


❒ Fig 10.5

Effect of cotyledon on seedling formation of Cassytha filiformis (after Rangaswamy and Rangan, 1971)

(viii) It has proved difficult to germinate by conventional methods the seeds of obligate phanerogamic root and stem parasites in the absence of contact with the host plant. Seeds and embryos of several obligate parasites have been grown under aseptic conditions to study their dependence on the host.

In 1962 Johri and Bajaj successfully cultured the young embryo of Cuscuta reflexa, a stem parasite in absence of host tissue (Fig 10.6). The mature embryo of Cuscuta has a definite plumule but it lacks a radicle. Truscott (1966) cultured mature and immature embryos of Cuscuta in order to induce root system by applying adenine, kinetin, GA_3, coconut milk and casein hydrolysate. But none of the treatments induced rooting. The experiment indicates that Cuscuta lacks a root growth potential.



❒ Fig 10.6

Culture of embryos of Cascuta reflexa

Undifferentiated embryo (0.5-1 mm) of Cuscuta reflexa on culture produce callus tissue which later form many embryoids. But differentiated embryos (1.5- 2.5 mm) produce seedling and later several adventives embryoids are formed.

In 1963, Rangaswamy and Rangan cultured the embryo of Cassytha filiformis, a stem parasite in absence of host. Root parasites are dependent on the host stimulus. Some root parasites e.g. Cistanche, Orobanche, Striga etc. are also dependent on the host plant for seed germination. In cultures the seeds of root parasites do not germinate in a nutrient medium. However, the addition of extract of host roots to the medium induces the seed germination independent of the host.

In vitro seed germination and shoot development independent of host stimulus has also been reported in root parasites Osyris wightiana, Exocarpus cupressiformis and Santalum album. Most of the root parasites do not form haustoria in cultures without a host.

(ix) The effect of nutrient medium, amino acids, vitamins and plant hormones can be observed on embryo in culture.

2. Applied Aspects of Embryo Culture:

From a practical point of view, in vitro embryo culture is very useful in many applied aspect:

(a) To raise a healthy plant from abortive or non-viable embryos;
(b) To overcome seed dormancy;
(c) To shortening the breeding cycle;
(d) To overcome self-sterility of seeds;
(e) Seed testing.

Culture of Non-Viable or Abortive Embryo:

Embryo abortion or non-viable embryos may develop in some natural seeds or may produce in seeds due to unsuccessful interspecific and inter-generic crosses.

Culture of abortive embryo from normal plant:

In the seeds of some fruit plants like apple, peach, pear etc. are abortive embryos which do not even respond to stratification treatment. In such cases, embryo culture is the most promising method to obtain a plant from such abortive embryos.

In the polyembryonic seeds like lemons, oranges, the additional embryos are developed from nucellar tissue. Such embryos gradually abort during development and do not germinate under natural conditions. These embryos can be grown under artificial in vitro conditions to mature plant for clonal propagation. When the goal is to obtain plants from abortive embryos, the embryos should be excised prior to the onset of abortion.

Culture of abortive hybrid embryo:

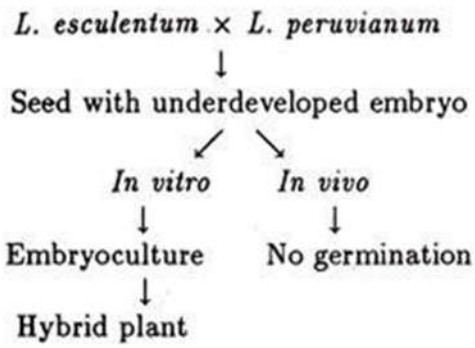
In many breeding experiments, embryo abortion is noticed in the seeds of unsuccessful crosses Although fertilization takes place normally in such crosses and hybrid embryos begin to develop in a relatively normal way, a number of irregularities subsequently set in which results in the formation of non-viable hybrid embryos.

As a result it makes a barrier to cross ability in plants where the non-viable hybrid embryos are unable to develop into mature plants. Plant embryo culture has now been used to speed up breeding programme and to overcome the cross ability barrier in plants. In breeding work with crop plant and horticultural plant, embryo culture method is very useful for the production of hybrid plants with desirable characters.

Crop Plants:

Embryo culture has been successfully employed in many unsuccessful interspecific crosses of crop plants. Tomato Cultivated tomato (Lycopersicon esculentum) is very susceptible to virus, mold and nematodes. But the wild species L. peruvianum is relatively resistance. In a

cross between the two species, the fruit develops normally but the seeds containing underdeveloped embryos do not germinate. Hybrid plants have been raised from such seeds by embryo culture.



Seed Dormancy and Embryo Culture:

Seed dormancy is a natural phenomenon Dormant seeds fails to germinate under apparently suitable conditions. In dormant seed the enclosed embryo is not able to grow. This temporary suspension of growth is definitely due to some internal condition of seeds or due to some environmental causes. Prolonged seed dormancy presents a special problem to plant growers.

So several methods have been devised to break the dormancy of seeds. But in some seeds dormancy cannot be broken by any conventional method Embryo culture has been successfully used to bypass the traditional treatments to overcome seed dormancy and accelerate germination in certain types of seed.

Seeds of tall bearded Iris remain dormant varying from a few months to many years after harvest. But within two to three months, transplantable plants are possible to raise by embryo culture. Seeds dormancy has been overcome in the seeds of some other plants such as Lactuca sativa, Citrullus colocynthis, Phacelia tenacetifolia, Nemophia insignis etc.

Seeds of some of the common cereals become dormant immediately after harvest. In vitro culture of excised embryos from such dormant seeds in suitable nutrient medium supplemented with hormone might be advantageously used to raise the seedling immediately after harvest.

In orchids, morphological development of the embryo and their subsequent germination take place in the soil in association with mycorrhizal fungi. Otherwise the seeds remain dormant in absence of symbiotic fungus. The seeds or odchid can be grown asymbiotically by culturing them in nutrient medium. Orchid growers are now commercially utilizing this technique for the production of profuse orchid plantlets.

Shortening of the Breeding Cycle:
In breeding practice, dormant seeds and slow growth of seedling are not suitable material. Embryo culture is a valuable tool to plant breeders because this method can reduce the breeding cycle.

Overcoming Self-sterility of Seeds:
In some economically important plants like banana, kachoo, seeds are produced, but such seeds are never known to germinate in nature. Such plants propagate very easily by vegetative means. This natural sterility barrier in the seed could be overcome by embryo culture.

Seed Testing:
Embryo culture has provided a rapid means of measuring accurately the viability and germ-inability of a particular lot of seeds. In seed testing practices, it is a very useful and reliable method to predict the viability of seeds.

3. Other Applications:
To Study the Evolutionary Relationship:
Conventional hybridization technique is an important method for the study of evolutionary relationship, speciation and for determining the taxonomic position of plants. In some cases hybridization fails at the interspecific level due to formation of abortive embryos. Hybridizing the different species of Datura and culturing abortive embryos in some cases, some interesting results has been obtained about their controvert taxonomic position and evolutionary relationship.

To Study Host Parasite Interaction:
Plant pathogens may infect the flower or flowering spike of host plant e.g. ergot disease of rye plant. This may lead to malformation o seeds or loss of seed. Incorporation of fungal of in the culture medium and the culture of embryo facilitate to study the host parasite interaction.

To Study the Mutagenic Effect:
Seeds are often irradiated by X-rays, and gamma-rays to study the mutagenic effect. Embryo culture is a very useful technique to evaluate" the mutagenic effect of the irradiated seed. From the practical point of view, embryo culture is also useful to determine the safety doses of irradiation used for food preservation.