# Artificial Seeds in Micropropagation

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## DEFINITION OF SYNTHETIC SEEDS

In the broadest possible sense, artificial seeds are a way of transferring somatic embryos or shoots from sterile tissue culture to nonsterile conditions, with or without an artificial seed endosperm. In a strict sense it is a somatic embryo with an artificial endosperm or seed coat. Thus, artificial seeds have no direct relation to the propagation method.

Redenbaugh et al. (1991) define the following four types of artificial seed:

- Uncoated, desiccated synthetic seeds
- Coated, desiccated synthetic seeds
- Coated, hydrated synthetic seeds
- Uncoated, hydrated synthetic seeds

The major research effort has been conducted with coated, hydrated synthetic seeds.

Encapsulation has several advantages over traditional acclimatization and soil establishment of somatic embryos.

1) Micropropagated plants can be delivered directly to the nursery/greenhouse without acclimatization. Acclimatization and several handling steps are saved.

2) The artificial endosperm and seed coat protect the embryo during transport, storage, handling, and sowing.

3) The small size of the propagule has advantages in distribution, storage, and transport.

4) You can supply the artificial endosperm with substances such as nutrients for use during germination, hormones that regulate germination, beneficial microorganisms, fungicides, and herbicides.

Consequently, artificial seeds can be used as an alternative to vegetative propagation for cloning of selected individuals or for propagation of costly and scarce hybrid seeds.

The major drawback of artificial-seed technology is cost. This is related to the fact that all work has been carried out at a research and development level. To my knowledge, artificial seeds have not been used on a commercial scale with any species. Apart from prohibitively high costs, the use is limited to plant species with a well functioning system for micropropagation by somatic embryogenesis. Therefore, for most commercially important plant species, improvement in micropropagation protocols are needed before artificial seed technology is possible. An additional disadvantage has been the low desiccation tolerance of the encapsulated hydrated seeds. To prevent desiccation it has been necessary to cover the somatic embryos with a polymer. However, this requirement has led to an oxygen deficit within the artificial seeds and a short storage life of the product.

## EXAMPLES OF ARTIFICIAL SEEDS

The species that one could imagine using for artificial seed production are crops with a high value such as cut-flower species, ornamentals, and vegetables. In addition, artificial seeds can be integrated into breeding programs for clonal propagation of hybrid seeds, transgenic plants, or other highly bred plants (Redenbaugh et al., 1988). To be able to use artificial-seed technology with a plant species there is a requirement for a commercially feasible micropropagation system. Table 1 shows the species where germination of artificial seeds has been achieved aseptically on nutrient media in petri dishes and under nonsterile conditions in soil (Redenbaugh et al., 1991).

**Table 1.** Species with artificial seeds.

| Aseptic germination in petri dishes | |
|---|---|
| Celery | Corn |
| Cabbage | Sweet potato |
| Carrot | Eggplant |
| Cotton | Eucalyptus |
| Alfalfa | Norway spruce |
| Rice | Mulberry |
| Lettuce | Sandalwood |
| **Germination in soil under nonsterile conditions** | |
| Celery | |
| Carrot | |
| Alfalfa | |

Alfalfa has been used as a model species. This is primarily because of the well developed micropropagation system and because alfalfa has exalbuminous seeds—seeds without an endosperm. Therefore, the nutrients necessary for germination are present in the embryo itself. The potential use of synthetic seed technology for alfalfa has been to clone hybrid seed and make them available to farmers. In this way, the time necessary for propagation of the hybrid seed can be shortened considerably while the genetic superiority of the hybrid seeds is maintained (Redenbaugh et al. 1988).

Mature somatic embryos of alfalfa have been encapsulated in an artificial endosperm of alginate and coated with a polymer to reduce the tackiness of and water losses from the artificial seed. This allows the artificial seeds to be handled like normal seeds and be sown mechanically. Field trials have shown that the artificial seeds need to be protected by a plastic polymer to obtain a reasonable frequency of embryo to plant conversion. Naked, pregerminated embryos have also been transferred to field conditions and show at least the same survival as artificial seeds (Fujii et al., 1992). In an experiment with automated encapsulation of somatic embryos the costs of artificial seeds have been estimated at 7¢ per artificial seed and 56¢ per plant in the field produced in this way (Redenbaugh et al., 1991). Further reductions in costs can be envisaged with commercial-scale artificial-seed production. Still, these prices are very high compared to the price of alfalfa seed, but the prices can be considered typical for a plant species with a micropropagation system of comparable quality. Bearing this in mind, the time when artificial seeds can compete with normal seeds is close for high value crops.

In the Botanic Garden at the University of Copenhagen, we have been working with somatic embryogenesis in *Picea abies* (Norway spruce), *Picea sitchensis* (Sitka spruce), and *Abies nordmanniana* (Caucasian fir or Nordmann's fir). In both spruce species the mature somatic embryos we can produce germinate and develop into plants at reasonably high frequencies. We have tried to encapsulate mature somatic embryos of Sitka spruce but have only achieved sporadic conversion. However, a recent report showed a 90% germination frequency of Norway spruce artificial seed under aseptic conditions (Fourré et al., 1991).

In Nordmann's fir, which is used for Christmas tree production, the *micropropagation system is not of the same quality.* We are only able to regenerate a low number of mature somatic embryos per petri dish and the germination capacity is not very high. On the other hand, Nordmann's fir is a high value crop in which the interest in using artificial seeds is very high.

## CONCLUSIONS

During the past years there has been a marked improvement in the micropropagation systems in a number of plant species and the quality of mature somatic embryos is getting closer to that of zygotic embryos. In addition, considerable progress in the desiccation tolerance of mature somatic embryos has been obtained and this opens possibilities for production of artificial seeds which can be dried and stored. Unfortunately, most research with artificial seeds has been done with embryos that are not dormant, do not tolerate desiccation, and cannot be stored. Therefore, the storage life of the product (the artificial seed) is very short and usability limited.

The ideal product is an encapsulated embryo, that can be stored in a dormant stage. Whether or not the embryo is desiccated is of minor importance. The major point is that it has some storage life. Apart from being dormant the artificial seeds also should have a high germination frequency—even after storage—in the field or greenhouse.

## LITERATURE CITED

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