# STORAGE AND MANIPULATION OF PLANT PROPAGATION MATERIAL

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Most of the published information on the storage of propagation material concerns species with a well defined dormant period. Deciduous perennial species are relatively easy to store. Dormant, bare root material can be kept in cold storage for up to two years. Desiccation is a potential problem and jacketed rooms are used to control this (7). There is also much information available on storage and manipulation of dormant bulbs. However, there is little information on the storage of actively growing vegetative material. Development of techniques to store this material could be of great benefit and will be the major consideration in this paper. The propagation industry is becoming increasingly sophisticated and uses involved techniques such as tissue and meristem culture and propagation from "virus free" clones. As a result there is an increasing need to be able to store and to transport actively growing plant material. It is essential to get this material from the specialised propagation units to the production nurseries without deterioration.

Although there is relatively little direct information available on propagation materials, we can learn much from existing technology for storage of vegetables and fruit. Modern storage and transport techniques are revolutionising the marketing of these commodities by enabling a continuity of supply of high quality product. Similar benefits should be possible for plant propagators.

The main aim of storage is to avoid change or to slow deterioration. Deterioration may take the form of continued growth and development, water loss, senescence, pathogenic infection, or attack by pests. All these forms of deterioration are slowed by reduced temperatures. At first it may seem strange to think of continued growth as deterioration. In most cases the freshly harvested propagation material is in the best condition and any change is undesirable. Lowering the temperature slows the normal metabolic and growth processes. The temperature used and duration of refrigeration that can be tolerated without causing damage will vary with different plant materials.

Senescence is the term used to describe the general degradation and ultimate death of living tissue. It includes yellowing and abscission of leaves and inability of the plant material to grow. Harvesting vegetative propagating material involves injury to the plant, removes its source of water and nutrients, and

greatly accelerates senescence. Storage or transport usually involves prolonged periods of darkness which again accelerates senescence. Lowering the temperature slows the senescence and therefore prolongs the useful life of the plant material.

Similarly, the activity of pests and diseases is generally reduced at lowered temperatures. However, there are exceptions to this and some pathogens may actively be stimulated at the low temperature. So there may be need for other pathogen control techniques as well.

Another factor in deterioration is loss of water. Vegetative cuttings are particularly susceptible to water loss. Their normal source of water, the root system, has been removed. Care must be taken to ensure a continual supply of water to the cuttings or to prevent the loss of water from them. Providing a continuous water supply during storage or transport can be cumbersome and messy. It is generally more practicable to prevent loss by enclosing the material inside a vapour barrier. However, there are complications with this also. The high humidity inside the container encourages pathogens and the composition of the atmosphere around the plant becomes modified. This aspect will be considered in more detail later. Once again, lowering the temperature can greatly reduce the rate of water loss but only if high humidity is maintained in the cool store. The conventional cool store is in fact a very effective dessicating system because the cooling coils are as much as 6°C below the required temperature of the room. Under these conditions there is extensive condensation and freezing of water onto the cooling coils. This dries out the atmosphere in the store and so increases the loss of water from the plants to the atmosphere.

Another factor is air movement. Forced circulation of the cooled air removes the high humidity air from around the plants, replacing it with the dried out air off the cooling coils. Dufresne (6) describes the jacketed room technique that is used to avoid drying out of propagating material. This is essentially a room within a room. Air is circulated over the cooling coils and through the space between the inner and outer rooms. So the walls and ceilings of the inner room become a very large cooling surface for the inner storage space. With this large cooling surface the temperature difference between it and the desired room temperature can be very small. Then there is little condensation onto the cooling surface and high humidity is maintained in the chamber. There is also no air movement in the inner chamber to hasten water loss. Rigid construction of this type of store is expensive.

An alternative is to use a conventional store and to enclose individual units or packages in plastic bags. On a large scale this is impracticable and uneconomic, and jacketed rooms were developed to avoid this problem (7). However, the C.S I.R.O has de-

veloped a system for converting conventional apple cool stores into controlled atmosphere stores by suspending a plastic tent inside the room. This is essentially a jacketed room and should be effective at a much lower cost. It should be a useful system for converting part or the whole of the storage space in existing cool stores into effective units for prolonged storage of propagation material.

Fruit and vegetables are frequently waxed to reduce water loss and to give an attractive gloss to the product. Perhaps there is some scope for using spray-on wax preparations on propagation materials. It would be necessary to ensure that the solvents used are not toxic to plants. The use of growth regulating compounds, particularly abscisic acid (ABA) may also hold promise. This has been shown to close the stomates on plant leaves and therefore greatly reduce transpiration. Certain analogues of ABA have been shown to close the stomates without inhibiting growth appreciably. The effect of one surface application lasted for 16 hours in Xanthium and for more than 9 days in young barley plants (8). It has been considered as a potential treatment to eliminate the need for mist in propagation of cuttings. Liitle and Eidt (10) showed that ABA reduced transpiration in hardwood cuttings.

In certain species low temperature may bring about desirable changes in the plant material. In Irish potatoes stored at low temperatures there is a change of starch into sugars. By putting them back to higher temperature the sugars revert to starch again (14). Perhaps it is possible to get better rooting of some cuttings after storing them at low temperature and so providing more sugars for the rooting process. However, each species will need to be tested because in some plants, e.g. sweet potato, sugars accumulate at high rather than at low temperatures.

Although refrigeration is the most important means of reducing deterioration, the effects of it are not all good. The most important disadvantage is chilling injury. This is damage to plant material by temperatures above freezing point (11). Not all plants are susceptible, and prolonged exposure is usually required to cause injury. A generalisation is that plants of tropical and subtropical origin are susceptible, while those from cool temperate regions are not. There are no specific symptoms of chilling injury, but it is expressed in different ways in different plants. Common sorts of symptoms are necrotic lesions or spots over the tissue and discolouration around veins. Lesions are frequently sunken because there is localised water loss from damaged cuticle and collapse of the cells beneath these damaged areas. In other cases the damage is present but the lesions are not visible. However, these damaged cells are very much more susceptible to pathogenic infection and the only visible symptom therefore is

excessive fungal and bacterial rots. An even more subtle form of chilling injury is only expressed as reduced capacity for growth. This type of injury can occur in early season field grown crops like beans or tomatoes, as well as in material exposed to low temperatures from mechanical refrigeration. The temperature and the duration of exposure influence the severity of the chilling injury. This type of injury where there are no specific symptoms but just reduced growth is extremely difficult to recognise. It is therefore imperative that propagating material is thoroughly tested for susceptibility to chilling injury before prolonged cool storage is attempted. There is evidence available to show that mild chilling injury in some species can be reversed by heat treatment (11).

Apparently chilling causes a change in the character of membranes in the plant cells so that normal volatile products cannot diffuse out. These then accumulate to toxic levels within the cells. A short heat treatment can revert the membranes to their normal state and allow the accumulated gases to escape. So it still may be possible and desirable to cool store susceptible material and briefly expose it to high temperatures at defined intervals to avoid chilling injury.

Another gaseous product of plant material is ethylene. This is, in fact, an important plant growth regulator (1). It is commonly used to induce ripening in fruits, particularly bananas. Ethylene is a normal product of plant metabolism and influences many aspects of plant growth. Although it is generally thought of as an inductant of ripening and senescence processes, there is now evidence for stimulation of root growth, fruit growth, germination and of elongation of cereals (9, 12, 16, 17, 18). However, the majority of known responses by harvested material are undesirable. It induces abscission of leaves, opening and discolouration of rose flowers, sleepiness in carnations and inhibits cell division (5). As with other growth regulating substances it is possible to find different plants which respond in opposite ways to the same compound. For propagation materials ethylene would be an undesirable compound, particularly in transport situations, as it speeds up the senescence processes. In cool storage it is not so serious as plants show less response at low temperatures. However, ethylene is a very common substance and handling plants, changing their orientation, and particularly cutting or injuring them induces very high rates of ethylene production. In preparing vegetative propagation materials there is extensive injury and therefore high rates of ethylene production. In the open air this gaseous ehtylene escapes quickly and so has little or no effect. If cuttings were prepared for transportation to distant locations, they would be enclosed in a sealed container and the ethylene could accumulate. This is most undesirable and

means of removing the ethylene should be considered. One convenient way is with potassium permanganate.

Scott (15) has developed a system for shipping bananas by enclosing them in a plastic bag with an absorbent block (vermiculite and cement) soaked in potassium permanganate. The permanganate inactivates ethylene produced by the bananas. Enclosing the fruit in a bag also results in modification of other gaseous components of the atmosphere. As the plant material continues to respire, it uses up oxygen and releases carbon dioxide. Both these changes depress the rate of respiration and therefore slows down the overall metabolism of the plant material, as does refrigeration.

Refrigeration is normally used to slow metabolism and so to extend the life of plant material. The same extension of life can be obtained with bananas by using refrigeration or the potassium permanganate block. I am not aware of this system having been used for propagation materials, but it could have application where they are stored or transported over periods of several days. If, as in the case of bananas, it is as effective as refrigeration, it should be a more economic practice. However, this sort of success is only possible if ethylene is an important cause of deterioration, as is the case with shipping of bananas or carnation flowers (3, 15).

Whenever living plant material is enclosed, whether in a plastic bag or in a sealed cold room, the atmosphere is modified by the respiration. We can deliberately alter the oxygen and carbon dioxide concentrations by absorbing or adding these as desired. This is known as controlled atmosphere storage and is highly successful with commodities like apples. Different plant material can tolerate different concentration of these gases without inducing damage. There is scope for testing controlled atmosphere storage with propagating materials. It may help to control specific storage disorders and to enable much longer storage.

Finally, I would like to mention a very interesting development by Burg in Florida (4). It is known as hypobaric storage and involves storage under a partial vacuum. This has three important effects. Firstly, reducing the pressure creates a refrigeration system. At the reduced pressure water evaporates and the energy required for this comes from the heat energy of the product. So, as water vaporises the temperature falls. The temperature reached is dependent on the extent of the vacuum. Typically, the pressure is reduced to one-fifth of an atmosphere and this would result in a temperature of approximately 20°C. It should be noted that this temperature reduction is only achieved by the evaporation of water. Unless free water is available, it will vaporise from the plant material and eventually cause wilting. This is avoided by

bleeding in humidified air or by injecting water vapour into the chamber.

The second effect is modification of the atmosphere. If the pressure is reduced to one-fifth of an atmosphere, the partial pressure of all component gases will be reduced to one-fifth of the normal concentration. Therefore the oxygen concentration will be reduced from approximately 20 % to 4 %, and will consequently reduce respiration rate.

The third and most important effect is the removal of ethylene. At the reduced pressure gases diffuse much more rapidly. Ethylene and other gaseous products of plant metabolism diffuse out of the tissue so rapidly that they do not have their normal effects. Ethylene is produced by all plant tissues and especially by injured tissue, and it stimulates respiration and senescence. A technique like hypobaric storage therefore has great potential for slowing deterioration of propagation materials.

Burg has shown that difficult-to-store cultivars of chrysanthemums can be stored for at least six weeks in hypobaric conditions. In normal storage they only last for 10 to 14 days. After the six weeks in hypobaric storage the cuttings were kept in normal storage for 10 days and still produced new plantlets as well as the fresh material. Other lines of chrysanthemum cuttings were kept in hypobaric storage for 12 weeks, then put in rooting beds for 2 weeks. After this treatment they grew perfectly and developed normal flowers in the usual time. This system seems to virtually suspend activity of these cuttings and offers great promise for the nursery industry.

JIM WELLS: Could you tell us precisely how those blocks are made with the potassium permanganate?

D. SIMONS: I think the proportions are one part cement to three parts vermiculite. Detailed information is available from C.S.I.R.O. Division of Food Science or from Departments of Agriculture. The block is merely a carrier — a material that will absorb a lot of liquid. It is important that the potassium permanganate not come into direct contact with the plant material as it is quite toxic. The blocks can be cut to any size to suit the container and the amount of ethylene expected.

VOICE: How would sponge plastic work?

D. SIMONS: It may be of some use, but the water holding capacity is much less than that of vermiculite. When sponge plastic is wet, it drains for a long time; vermiculite doesn't drain so much. Any carrier will work, or even free liquid potassium permanganate, so long as you avoid contact with the plant.

ARNOLD TEASE: In sending cuttings from one country to another could this have practical application?

D. SIMONS: Certainly. It should have application anywhere cuttings are kept in a closed space.

VOICE: Is literature available on the low pressure system?

D. SIMONS: Yes. The most recent information is in the article by S. P. Burg.

VOICE: You mentioned the use of abscisic acid sprayed on cuttings. Are there references to this?

D. SIMONS: There are numerous references to the effects of applied abscisic acid but I know of none in the specific context mentioned. I suggested it as one of the possibilities. References (8, 10) may be useful.

KEN TURNINGS: Is it true that cuttings taken from plants whose foliage normally hangs pendulous and then turned into an upright position would give off more ethylene than cuttings taken from a plant which normally stands erect?

D. SIMONS: I can't really answer that because I don't know of it having been tested. Work done about 40 years ago showed that if asparagus spears were placed horizontally, they produced more ethylene than if they were in the vertical position. Tomato fruits placed with their stem scar down ripen more quickly than those in the normal position of stem up. Rotating plants in a klinostat or movement in the wind also induces ethylene production. It appears that changing the orientation of plants induces ethylene production and I would expect the same to apply in the situation you proposed.

JOHN OAKLEY: You mentioned that storing cuttings at different temperatures could change the conversion of starch to sugars. How long would it take for this conversion to be effective?

D. SIMONS: This would vary with the temperatures used and the plant material. In very general terms it is a slow change, particularly at low temperatures. In potato, where this has been thoroughly investigated, the change from starch to sugar occurs slowly over several weeks. If this sort of change is of any significance in cuttings, I would expect it only to occur slowly over prolonged storage periods. To my knowledge it has not been tested in cuttings.

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