Seed Notes
for Western Australia

No. 3 Seed quality and germination

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This issue of **Seed Notes** will cover some simple methods to determine the quality of your seed and provide basic information on germination testing.

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- Determining seed quality through visial assessment, flotation methods and sectioning
- Seed germination techniques for canopy stored seed, hard seeded species, small seeded myrtaceaous species and for some difficult species
- Recommended reading

Seed quality and germination

To grow plants from seed, some knowledge of the quality and germination requirements of your seed is required. Not all fruits will produce seed. Not all seed produced will be viable or germinable. Seed is a highly variable resource and lack of seed production in flowering plants can be caused by a number of factors. These include pollination failure, resource deficiency, predation and genetic defects causing developmental failure. Environmental stress and age structure of the population can also contribute to a low seed production.

The first leaves appearing after germination in seed of Hemigenia.
Photo – Anne Cochrane

Above left: Germinating seed of Villarsia *sp.*
Left: Hibbertia *seed germinating after seed coat removal.*
Photos – Anne Cochrane

Cost effective and simple methods to identify seed quality and germinability are required for any seed related venture. The techniques outlined below incorporate some general principles for assessing seed quality and germinability. They are not definitive. It is important to remember that there is great variability in seed production between plants, sites and years of collection. Your results for one seed batch are unlikely to be the same for another seed batch. Each collection must be assessed on an individual basis. You may discover other methods for assessing quality and germinability of your seed that are not listed, but that work well for your species. If some techniques work well, share your knowledge and we will all be the better for it.

Seed quality testing

There are a number of quick tests that can be conducted on seed to determine whether or not the seed is potentially viable. Insect damaged seed and shrivelled seed is unlikely to germinate. Fruits with no seed within will also not germinate. Don't waste your time on trying to germinate, store or direct seed material that is no good. Spend some time on assessing seed quality using one or several of the methods below.

Dryandra seeds.
Photo – Anne Cochrane

Shrivelled seed of a Gastrolobium.

Plump, healthy seed of a Daviesia.
Photos – Anne Cochrane

REMEMBER:

Always assess your seed in some way. If at a later date the success of your planting effort was dismal, it may have been due to lack of germinable seed within the seed batch used. Avoid disappointment and do quality assessments and/or germination tests.

Visual inspection

Very simply, look at your seed closely. If your seed is large use the naked eye. Use a magnifying glass or microscope if the seed is very small. Do some seeds appear shrivelled? Do some seeds have holes in them or frass (the debris or excrement of insects or insect larvae, which can look like sawdust) around them? These signs may indicate insect damage. Are the seeds broken or damaged in any way? Too vigorous cleaning of the seed may have caused some damage to the outside coat and also the inside of the seed itself. The seed that is healthiest and most likely to germinate will be undamaged and plump. After a while you will get a feel for the look of the seed and you should be able to discard any seed that does not look full, undamaged and plump. Often green seed is a sign of immaturity and these seeds may not germinate because of incomplete development. It is usually fairly easy to determine whether the seed of species with canopy-stored seed (e.g. *Banksia*, *Hakea* and *Dryandra*) is healthy. Once the seed is extracted, check to see if the seedcoat is dark brown or black and the inside of the seed is firm and white. The hard seed of genera in the pea families can also be assessed visually if not too small. Any insect damaged and shrivelled seed should be discarded. In some cases you will not be able to tell a good filled seed from an empty one as many species produce 'dummy' seed—a fully formed hard seedcoat with nothing within. This has been known to occur in the genus *Adenanthos*.

Darwinia *seeds. Note the difference between the fruit on the right (swollen with seed) and on the left (shrivelled and not containing a seed).*
Photo – Anne Cochrane

Seed sectioning (cut test)

The cut test is a very simple method to determine whether your seed is full (contains an endosperm, the food necessary for growth and development) or empty. Simply take a representative sample of your seed, either by weight or by a count, and cut each seed carefully to see if there is anything inside. Generally, a good healthy seed will be firm and white inside, not shrivelled or overly dry. Empty fruits will be very obvious and may even have evidence of insect damage or an aborted seed. Often the fruit will be very plump and it will be obvious that there is likely to be a good seed within; in other cases the fruit will appear shrivelled in comparison to the plump one. Cut them both open and you should see the difference immediately.

Many *Eremophila* species produce few or no seed within their hard woody fruits. Research into seed set in *Eremophila* at the Threatened Flora Seed Centre has shown that not all fruits contain seeds and in many cases less than 50 per cent of fruit are filled. The many compartments or locules of the fruit are not visible in undamaged whole fruits, and an assessment of number of seeds per fruit is therefore difficult without opening a representative sample of the fruits. Some *Eremophila* fruits are very hard and sectioning with a scalpel is difficult. The seed is often quite small and a microscope may be necessary for this job. But in general it is feasible to do a cut test on these fruits.

Bissected Eremophila *fruit showing aborted seed*
Photo – Anne Cochrane

Cut tests on *Allocasuarina* can be very useful to identify full from empty seeds. It is also possible to use this technique of assessing seed quality on the seed of *Adenanthos*, *Petrophile* and many other species that produce single-seeded fruits.

In *Verticordia*, *Darwinia* and *Chamelaucium* the small seed is a nut enclosed within the old flower receptacle. Not all receptacles contain a fully formed seed. Seed set in *Verticordia* may be anything from zero to 68 per cent, with much variation between plants, collection sites and years of collection. Five grams of material may represent many thousands of flowers but the number of seeds held within that weight may be in the order of 100 or less. A cut test on these species will determine how much seed has set per unit weight. Ideally do your cut test on several replicates of 50 or 100 fruits to ensure that you are getting an average over the whole collection.

A small section of fruit wall cut away to reveal healthy endosperm.
Above: Darwinia.
Below: Conospermum.
Photos – Anne Cochrane

Reduced surface tension flotation test (float test)

This is an effective method to determine whether seed is full or empty. The test is simple and requires a container to hold water (a bucket, jar or cup depending on the quantity of seed being tested), a few drops of detergent to act as a surfactant, and your seed. Place the seed into the container with the detergent and agitate all together. Seed should either float or sink. Sinking seed should be full and healthy, while floating seed is likely to be empty or shrivelled.

Seeds of both Boronia *(above) and* Hemigenia *(right) can be subjected to a float test to determine whether seed is filled.*
Photos – Anne Cochrane

Do a cut test on a sample of each of the floaters and the sinkers to see if this is correct.

This test works well with hard-seeded peas in the family Fabaceae (e.g. *Daviesia*, *Chorizema*, *Gastrolobium* and *Gompholobium*) and Mimosaceae (e.g. *Acacia*), and has also been used on species in *Hemigenia* with good success. Do not attempt this test on seed of *Allocasuarina*. *Allocasuarina* seed is mucilaginous. This means it has a mucous membrane around the seed that gets very sticky on wetting. The seed clumps together and becomes a congealed mess. This is an adaptive mechanism that helps seed establish in micro-sites. In essence, it has its own wetting agent that will help germination and establishment.

Seed germination testing

Seed germination trials can be carried out in any type of container on filter paper, vermiculite or sponges, or some other medium that can be kept moist while the seed germinates. Agar, a jelly-like nutrient-free medium can be purchased as a powder and made up in distilled water (one gram per litre of water). Heat and stir the mixture on a hot plate until it clarifies. This should be just below boiling point. Pour the liquid into dishes when cooled sufficiently not to be dangerous to handle or to melt plastic containers. Any clear container with a lid should do for germination trials. Clear take-away containers such as hamburger containers can be used. Round plastic Petri dishes are ideal, cheap and readily available from laboratory suppliers. Otherwise use normal pots and seedling punnets. Most seeds require light, moisture and a temperature somewhere between 15 and 25˚C for optimal germination. Some seed requires a pre-treatment before germination will commence. This may be a heat or scarification treatment, a soaking in a smoked water solution, the use of more sophisticated growth hormones, or a combination of a range of treatments. In most cases the species will not require complex conditions for germination.

A temperature and light controlled incubation cabinet can be used to assess the germination capability of seed.
Photo – Michael James

Canopy-borne seed

Canopy stored seed held in woody fruits such as occurs in *Banksia*, *Hakea*, *Lambertia* and *Dryandra* tend to germinate readily without any pre-treatment once seed is extracted from the woody fruits. Light, moisture and a moderate temperature will suffice. Germination may take up to one month.

Dryandra *seed.*
Photo – Anne Cochrane

Hard-seeded species

Seed of pea species such as *Acacia*, *Daviesia*, *Gastrolobium* and *Chorizema* generally require cracking of the hard seed coat before germination will commence. The hard seed coat prevents the seed from taking up water. This cracking of the seed coat can be achieved by mechanical scarification (rub the seed gently over sandpaper or an emery board) or by pouring near boiling water onto the seed and allowing the seed to soak in the cooling water for two or more hours. It is also possible to crack the seed coat by nicking a small portion of the coat with a scalpel to reveal the seed beneath. The seed itself must not be damaged or it will die. This technique requires more skill and should be conducted with the aid of a dissecting microscope. Germination of seed of many hard-seeded species is relatively quick and may commence one week after treatment.

Germination of two hard seeded species after hot water treatment. Note radicle emergence.
Phtotos – Anne Cochrane

Small-seeded Myrtaceous species

These include seed from the genera *Eucalytus*, *Melaleuca*, *Callistemon* and *Kunzea* to name a few. The capsules of these plants contain not only seed but also chaff (sterile and aborted ovules). Chaff does not germinate, and it is often very difficult to distinguish between seed and chaff in some species. In other species such as *Eucalyptus calophylla* (marri) the seed and chaff are readily distinguishable. If you have access to a microscope the task of identifying seed will be easier. Otherwise it is necessary to weigh the collection of seed and chaff, take out a representative sample and place both seed and chaff onto a medium for germination. Most of these small seeded myrtaceous species contain no dormancy mechanisms to prevent their germination and only require light, moisture and a suitable temperature. In the case of these small-seeded species, the weighed replicate test (number of germinable seed per unit weight) may be the most accurate assessment of your seed batch.

Left: Eucalyptus macrocarpa *fruit.*
Photo – Anne Cochrane

Difficult species

In our experience there are a range of native species from the south-west of Western Australia that are in this category. They come from a range of families and include *Eremophila*, *Verticordia*, *Darwinia*, *Chamelaucium*, *Conospermum* and *Andersonia*. Many require the seed or fruit coat to be removed before germination can proceed. In the natural environment this will occur over time, after passage through an animal gut or through insect or micro-organism activity. This means that germination will be hard to replicate without some specialised equipment or a great deal of time waiting (from one to two years in some cases). For difficult species we have no easy recommendations. Many of these seed types respond to soaking in a smoke water solution for up to 24 hours before sowing. If access to a microscope is available, then carefully removing the seedcoat structures without damaging the seed, soaking the seed in smoked water and placing the seed in a container for germination should give some success. Seeds of these species often respond to applications of naturally occurring plant growth hormones such as gibberellic acid as the compound GA_3. This hormone can be purchased in powder form from chemical companies, such as Sigma, and is made up with a phosphate buffer to the desired strength (the stock solution). 1000ppm is recommended. A small amount of this mixture is then added to the medium, or else the seed can be soaked in a diluted stock solution for several hours before sowing. Around 25mg to 50mg of stock solution is added to a litre of water to stimulate germination.

Seed and chaft of a eucalypt—the seed is dark and the chaff is pale.
Photo – Anne Cochrane

For more information on seed quality assessment and germination the following references will be helpful:

Bell, D. T., Plummer, J. A. and Taylor, S. K. 1993. Seed germination ecology in south-west Australia. *The Botanical Review* 59, 24-73.

Bradbeer, J. W. 1988. *Seed Dormancy and Germination*. London, Blackie Academic and Professional.

Langkamp, P. 1987. *Germination of Australian Native Plants*. Inkata Press, Melbourne.

Mayer, A. M. and A. Poljakoff-Mayber 1989. *The Germination of Seeds*. Oxford, Pergamon Press.

Ralph, M. 1994. *Germination of Local Native Plant Seed*. Murray Ralph, Melbourne.

Ralph, M. 1993. *Growing Australian Native Plant Seed*. Murray Ralph/Bushland Horticulture, Melbourne.

For suppliers of growth hormones, germination media, filter papers and petrie dishes consult the Yellow Pages under laboratory equipment and/or suppliers.

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These **Seed Notes** aim to provide information on seed identification, collection, biology and germination for a wide range of seed types for Western Australian native species.

THREATENED FLORA SEED CENTRE

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