Germination testing: environmental factors and dormancy-breaking treatments

Technical Information Sheet 13b

For germination to occur, seeds require the correct combination of environmental conditions: moisture, temperature, light and gases. Suitable germination conditions are likely to be similar to those at the time of natural seedling emergence.

Specific **environmental cues**, such as light and alternating temperature, are often required by small-seeded species, which need to germinate close to the soil surface. Conversely, larger seeds, such as those that occur in coastal dune or desert habitats, which are at risk of drying out, may require darkness and constant temperatures for germination. Germination of some species (e.g. aquatics) can be promoted by anaerobic conditions.

Seeds that do not germinate under favourable conditions are considered dormant. Seed dormancy has evolved to ensure that germination is delayed until the environmental conditions for seedling growth and establishment are optimum. For example, temperature, light and moisture conditions may be suitable for seed germination twice in a year, e.g. in both autumn and spring, but chances of seedling survival are much lower if seedlings emerge too early. Therefore, a dormant period ensures that seed germination is delayed until the appropriate season (Fig. 1).

Types of seed dormancy

Several different classifications have been proposed to describe seed dormancy. The two most common are physical (seed coat related) and physiological (embryo related).

- **Physical dormancy** (PY), or exogenous dormancy, describes seeds which possess a hard seed coat that is impermeable to water (e.g. Fabaceae).
- **Physiological dormancy** (PD), or endogenous dormancy, refers to seeds in which the embryo possesses a physiological inhibiting mechanism that prevents radicle emergence (e.g. Amaryllidaceae).

Other dormancy types include:

- **Morphological dormancy** (MD) describes seeds with a small or underdeveloped embryo (e.g. Apiaceae).
- **Morphophysiological dormancy** (MPD) describes seeds with PD, which also have an underdeveloped embryo, that needs time to grow before germination can occur (e.g. Ranunculaceae).
- **Combinational dormancy** refers to seeds possessing both PY and PD (e.g. Rhamnaceae).
- Dormancy can also be classified as chemical, due to the presence of inhibitors in the pericarp (e.g. Oleaceae) or mechanical, due to the presence of a hard, woody fruit wall, usually an endocarp (e.g. Meliaceae).

Choosing germination conditions

The typical process for carrying out germination testing begins with research. Carry out a literature search for your plant species for storage behaviour and published test protocols. Use climate data such as WorldClim (Hijmans et al., 2005) to determine the optimum germination temperature for the species. Apply a dormancy breaking treatment if dormancy is suspected.

Check seeds at regular intervals for germination. Once germination stops, cut test any seeds which have not germinated (see Technical Information Sheet 13a).

When selecting germination conditions think about which natural environmental cues would stimulate the seed to germinate, and the conditions and dormancy breaking treatments required to overcome seed dormancy (see Box 2).

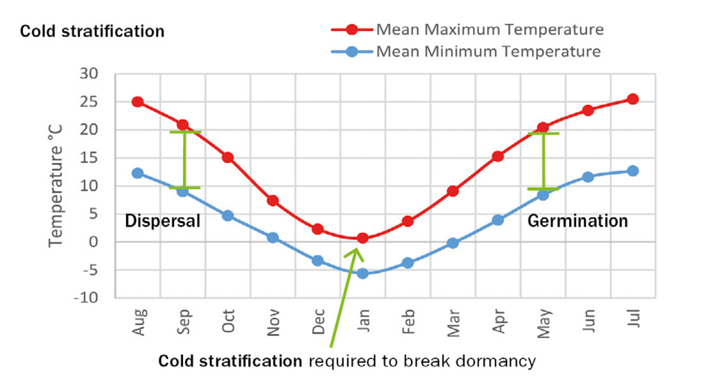

Figure 1: Average maximum and minimum monthly temperatures at the seed collection location of *Persicaria maculosa* in Slovakia. Dormancy delays germination until spring, despite similar environmental conditions in autumn. This species also requires a daily alternating temperature of 20/10°C, for germination.

Box 1: Germination requirements

Germination requirements are determined by an integration of:

What kind of plant

- Life form (tree / herb / annual / perennial)
- Taxonomy (family trends)

Habitat and climate

- Habitat (terrestrial / aquatic)
- Climate (temperate / tropical)

What kind of seed

- Endospermic / non-endospermic
- Nature of covering structures
- Position and size of embryo

Is seed dormancy expected?

Box 2: Germination and dormancy breaking considerations

Environmental cues

- light / dark
- constant / alternating
- temperatures
- aerobic / anaerobic conditions

Dormancy-breaking treatments

- cold / warm stratification
- dry after-ripening
- surgical treatment
- chemicals (e.g GA$_3$)
- scarification

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Dormancy breaking treatments

- **Scarification** (Fig. 2) alleviates PY by breaking the seed or fruit coat that prevents water uptake (imbibition). The coat is 'chipped' away from the root axis to avoid damaging the radicle. Alternatively dry heat (oven), wet heat (boiling water), or sulphuric acid may be used. These methods can be applied to larger seed samples, but risk damaging seeds.
- **Cold stratification** releases PD in imbibed seeds held at cold temperatures. This mimics the conditions a seed would experience over winter before germination in spring.

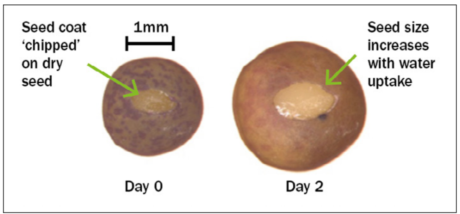

Figure 2: Scarification to break PY in *Vicia tetrasperma*. Remove a portion of the seed coat away from the root axis to allow water uptake.

- **Warm stratification** alleviates PD at warm temperatures, mimicking conditions through summer, for a seed that would germinate in autumn.

Several seasonal cycles of warm and cold stratification, of several months' duration, may be necessary to break dormancy in some species.

- **Dry after-ripening** mimics the conditions of a dry season required to break PD (Fig. 3). This technique is applied by placing seeds at intermediate humidity (*c.* 60% RH, using specified concentrations of LiCl solution), and warm temperatures (*c.* 20 – 30°C). See Technical Information Sheet 09 for the LiCl solution preparation protocol.
- **Surgical treatment** alleviates PD by removing a portion of tissue close to the root tip which is preventing radicle emergence. This can substitute for natural dormancy breaking treatments (e.g. stratification or dry after-ripening) at a mechanical level. The incision technique may need to be adapted depending on the embryo structure and position (Fig's. 4 - 6).

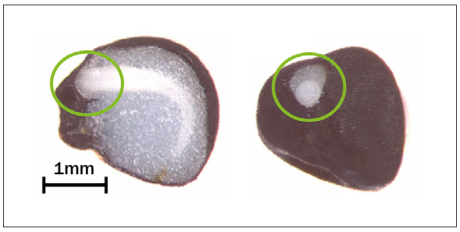

Figure 4: Surgical treatment to alleviate PD in *Allium neapolitanum*. Remove a small portion of the seed coat over the root axis.

Figure 5: Surgical treatment of *Eragrostis*. To release dormancy, remove a small portion of the seed coat along the distal ridge above the embryo.

- **Chemicals** such as gibberellic acid (e.g. GA_3), nitrates (e.g. KNO_3) and smoke can also be used to promote germination. They are typically applied by pre-soaking seeds or including the chemicals in the agar, see Technical Information Sheet 13a for more information on preparing GA_3 solutions. GA_3 can replace cold stratification in some species (e.g. Lamiaceae).

Box 3: The plasticity of dormancy

- A population of seeds will display a normal distribution of dormancy states, which can change through time.
- Just because a species usually displays a particular form of dormancy; non-dormant populations could exist.
- It is important to include controls for each dormancy breaking treatment.

Figure 6: Germination of *Eragrostis prolifera* after surgical treatment.

Equipment specifications

Refer to Technical Information Sheet 13a for information on suggested equipment for germination testing.

Acknowledgements

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Further reading

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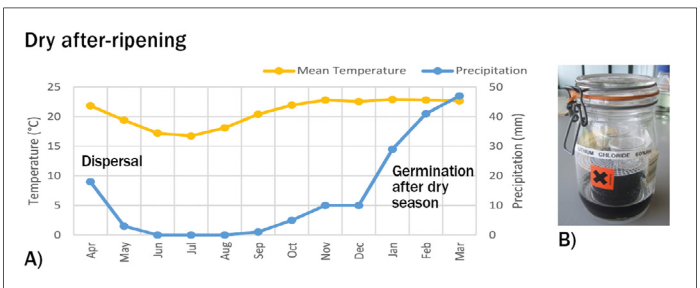

Figure 3a: Average monthly temperatures and rainfall at the *Eragrostis* seed collection location in Burkina Faso. Dormancy delays germination until after the dry season. Figure 3b: Seeds 'after-ripening' in the lab. Place seeds on a dry dish inside an air-tight jar containing a 60% relative humidity (RH) solution of lithium chloride (LiCl).