Measuring seed moisture status using a hygrometer

Reliable monitoring of seed moisture status is important in the handling and processing of seed collections:
• Seed collectors in the field need to know the moisture status of freshly collected seeds in order to handle collections properly (see Technical Information Sheet_04).
• Seed bank staff need to know when seeds are dry enough to store, and to identify collections in which drying may be slow.

This information sheet describes the method used by the MSB to provide a reliable, non-destructive measure of seed moisture status.

Procedure
Place a sample of seeds into the chamber of a hygrometer. Allow them to equilibrate and record the end point eRH and temperature.

Sample size
Ideally the seed sample should fill the chamber, or should occupy at least 20% of the total chamber volume. If you have a small sample of seeds, reduce the chamber volume using a metal insert.

Equilibration
Allow at least 30 minutes for seeds to reach equilibrium. Ideally, take periodic measurements or log the time to reach equilibrium.

The time taken for the seeds to reach equilibrium with the air in the sample chamber depends on:
• temperature of sample and sensor;
• initial water potential difference between seeds and air;
• whether seeds are wet or dry - wet seeds will equilibrate more quickly;
• whether seeds are in equilibrium with their environment prior to measurement - seeds not in equilibrium have a moisture gradient within their tissues (larger seeds have a steeper gradient);
• permeability of seed covering structures.

Species with impermeable seed coats (i.e. those with physical dormancy) present particular problems. These seeds have external structures that prevent the free movement of water, so will not reach equilibrium with their environment. To avoid making potentially false eRH measurements, seeds with impermeable coats need to be cut, coarsely ground or crushed before being placed in the sample chamber of a hygrometer. Work quickly to avoid changes in moisture status of the seeds. As seeds are destroyed in this process, the test becomes ‘destructive’.

Practical tips
• To avoid inaccurate readings, minimise direct handling of seeds, do not touch the inside surfaces of the sample chamber and do not breathe on an exposed sensor.
• Keep seals clean and check periodically that moisture cannot leak in or out of the sample chamber during measurement.
• Clean sample chambers with deionised water and/or alcohol and dry carefully. Some manufacturers supply sample pots to prevent contamination of the sample chamber.
• Check sensors regularly and keep them free of contamination. Follow manufacturer’s recommendations for cleaning.
• Calibrate hygrometers regularly (see overleaf) to maintain accuracy.

Sample Temperature

eRH values are temperature dependent. If there is a difference between the temperature of the seeds and the temperature of the hygrometer sample chamber, the eRH value recorded will be incorrect. If possible, take measurements under controlled conditions and always state the temperature at which the eRH measurement was taken, e.g. 15% eRH at 15°C. For routine eRH measurements, for example checking if seeds are dry enough to store, set the hygrometer up in the dry room.

Take particular care when measuring the eRH of seeds removed from the cold room. If very cold seeds are exposed to ambient temperature, even for a couple of minutes, water may be absorbed onto the surface of the seeds by condensation. This (additional) water would be quickly lost to the sample chamber atmosphere during measurement, giving an inaccurate, high reading. To avoid this, allow ample time for the collection to warm up to the measurement temperature, before opening the container and transferring seeds to the hygrometer sample chamber.

The method measures equilibrium relative humidity (eRH); that is, the relative humidity (RH) of air at equilibrium with seeds held in a sealed chamber. It is rapid, and non-destructive, making it ideal for small collections of rare and threatened species.

If required, eRH measurements of seed moisture status can be related to seed moisture content by reference to isotherms (see Technical Information Sheet_09).
Inexpensive alternatives
Acceptable estimates of seed eRH can be obtained from a range of less expensive hygrometers:
- Probe-type hygrometers
  Meteorological hygrometers can be used to measure seed eRH, provided that the probe (containing a sensor) can be inserted into a suitable ‘sample chamber’ through an air-tight seal.
- Self-contained data loggers
  When placed amongst seeds inside a cloth bag, data loggers can monitor changes in moisture status during transit and provide important data to explain the quality of collections arriving at a seed bank. Make sure that the sensor is completely buried within the seed collection.
- Low-cost mechanical and electronic hygrometers
  Mechanical or electronic hygrometers can be purchased relatively cheaply. Although they are less accurate, these instruments can be placed amongst seed collections during a field trip, to help indicate whether collections need to be dried. They can also be used to check whether ambient conditions are suitable for drying and to confirm that collections have dried.

See Technical Information Sheet 07 for further details on Low-technology monitors of seed moisture status.

For research applications, it may be necessary to warm or cool the sensor and chamber to the temperature of the seed sample before starting the measurement.

Field use of digital hygrometers
Many digital hygrometers are designed to operate by battery as well as mains power and can be used in the field. Measuring ambient RH (by exposing the sensor head of the hygrometer to the air) and seed eRH at the time of harvest can guide post-harvest handling decisions (see Technical Information Sheet 04) and provide useful data on seed moisture status at the point of dispersal.

For good results from in-field measurements:
- Keep the hygrometer in the shade to avoid warming the sample chamber and sensor. Avoid holding the sample chamber during measurement. If the chamber is warmer than the ambient temperature, seed eRH values will be too high and ambient RH values too low.
- Unless the collection is going to be split into distinct maturity classes for post-harvest handling, ensure that the seed sample used for eRH measurement is representative of the bulk.
- When taking eRH measurements to inform post-harvest handling, test the complete fruiting structure that has been collected.
- If seeds are going to be cleaned immediately after harvest, record the eRH of the extracted seeds.
- When taking eRH measurements to provide information on the seed moisture status at the point of dispersal, use only fully ripe seeds, extracted from the fruit. Unripe seeds or ‘green’ fruit tissue could give a false high reading.

Calibration
- Always use certified Lithium Chloride standards and carry out calibration at a constant temperature.
- Select calibration standards according to local conditions but always include a low (e.g. 0 or 5% eRH) and a high (e.g. 95% eRH) standard and 3 intermediate standards (e.g. 20, 50, 80% eRH).
- Using forceps, place one absorbent pad in a re-usable small plastic chamber.
- Carefully break open an ampoule of the selected calibration standard and drip the entire contents onto the absorbent pad.
- Place in the hygrometer sample chamber, seal as normal and leave to equilibrate (45min – 2hrs, depending on standard).
- Repeat for the remaining standards and compare results with expected eRH. Plotting the results should produce a straight line. In order to monitor long-term sensor behaviour (drift), do not adjust the sensor following calibration, but instead apply a correction factor based on the calibration.

Further reading

Equipment specifications

<table>
<thead>
<tr>
<th>Description</th>
<th>Model/Product</th>
<th>Supplier</th>
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<tbody>
<tr>
<td>Lab-based hygrometer with docking station and clamp</td>
<td>AW-DIO sensor with HygroPalm 3 display unit. Battery or mains operated. Range: +5 to +50°C (± 0.2°C); 0 to 100% RH (± 1.5% RH).</td>
<td>Rotronic Instruments (UK) Ltd. <a href="http://www.rotronic.com">www.rotronic.com</a></td>
</tr>
<tr>
<td>Portable relative humidity logger with mini probe</td>
<td>HygroClip SC04 sensor with HygroLog-D unit. Battery operated. Range: -40 to +100°C (± 0.3°C); 0 to 100% RH (± 1.5% RH).</td>
<td>Rotronic Instruments (UK) Ltd. <a href="http://www.rotronic.com">www.rotronic.com</a></td>
</tr>
<tr>
<td>Data loggers for measuring ambient relative humidity</td>
<td>Tiny Tag and Tiny View loggers. Range: -30 to +50°C (± 0.2°C); 0 to 100% RH (± 3% RH).</td>
<td>Gemini Data Loggers (UK) Ltd. <a href="http://www.geminidaloggers.com">www.geminidaloggers.com</a></td>
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Please note that the above equipment is used by the Millennium Seed Bank Project and has been chosen carefully using our many years’ experience. The list of suppliers is for guidance only and does not represent an endorsement by the Royal Botanic Gardens, Kew. The manufacturer’s instructions must be followed when using any of the equipment referred to in this Information Sheet.

Left: Hand-held Rotronic hygrometer with probe