A Water Activity-Regulated Dryer: How To Dry Seeds or Pollen With Water and No Heat

Patrick Baldet and Fabienne Colas

Research Engineer, Forest Genetic Resources Specialist, Irstea, Unité Écosystèmes forestiers, Nogent sur Vernisson, France; Researcher, Seed and Pollen Specialist, Ministère des Ressources naturelles, Direction de la recherche forestière, Québec City, Québec, Canada.

Abstract

The hydrous status of seeds and pollen can now be characterized more precisely than moisture content through the measurement of water activity (a...). This new technique, now available to managers of genetic resources banks, offers the advantage of being fast, easy to use and, above all, nondestructive. This article describes components and use of an a -regulated seed/pollen dryer developed by the National Research Institute of Science and Technology for Environment and Agriculture (Irstea, France). The simple design of this highly versatile heat-free dryer allows homogenous drying of seed and pollen lots at an a_w level that is safely compatible with reliable medium- and long-term conservation. The ex situ conservation of biodiversity is a major issue in the context of climate change. Hence, the dryer proves to be a high-performance tool in readjusting the hydrous status of lots likely to change during conservation.

Introduction

The measurement of water activity (a,), often associated with the measurement of equilibrium relative humidity (ERH), is a technique developed and widely used by the agrifood industry (Barbosa-Canovas and others 2007). This technique allows for characterizing interactions between water and other component molecules and predicting their potential for conservation. One property of hygrophilous materials is the ability to capture or yield water depending on the ERH of surrounding environmental conditions. At equilibrium, the a, qualifies the water status in the material, while the ERH qualifies the surrounding environment conditions. In more technical terms, the a is assessed by measuring the ERH on immediate contact with the sample; hence, the two measurements are often related. The a_w is the ratio between the water vapor pressure of a sample and the pressure of pure water, and ranges from 0 to 1. This ratio ranges from 0 to 100 percent when expressed in terms of ERH. This measurement technique is fast, reliable, reproducible, and nondestructive. In addition, operators only need basic training to measure a...

In general, the prestorage hydrous status of organic materials, referred to as intermediate humidity, is an influencing factor in their conservation and longevity. To date, the hydrous status of forest reproductive materials (seed and pollen) has been generally assessed by measuring mass moisture content. But this measurement technique is time consuming, destructive, and only quantitative. In addition, the chemical availability of water and its potential adverse effects on the conservation of stored lots cannot be qualified with this technique. Conversely, the measurement of a_w provides an accurate assessment of the deterioration risk related to water status (Baldet and others 2009).

Since 2004, Irstea (The National Research Institute of Science and Technology for Environment and Agriculture, France, formerly known as Cemagref) has demonstrated the usefulness of a_w in the forest genetic resources field through the application of this technique to the control of seed or pollen hydrous status (Baldet 2006). The degradation agents of seeds and pollen, whether biotic (bacteria and mold) or abiotic (oxidation and enzymatic reactions), are not dependent on the amount of mass moisture contained in a given compound, but on the chemical availability of water qualified by the measurement of a_w (figure 1). For instance, humans have known for thousands

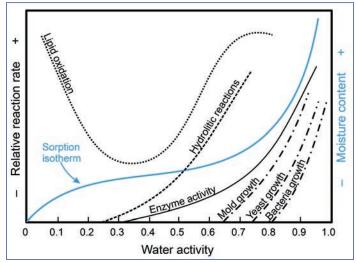


Figure 1. Schematic diagram of the main degradation agents of organic materials associated with the chemical availability of water. (Adapted from Labuza and others, 1972)

of years how to prolong the preservation of foods by adding "water binders" such as salt or sugar to reduce the fraction of chemically available water.

Through collaborative work, Irstea and the Direction de la recherche forestière (DRF) of the Ministry of Natural Resources (MRN) of Quebec determined the universal a value of 0.35 for the safe storage of orthodox seeds and pollen (that can be safely dried) by describing the hydrous behavior of hundreds of seed and pollen samples (Colas and others 2010). The a 0.35 value is a noteworthy value at which the main degradation factors are the least active (figure 1). Therefore, this value helps to prevent the development of any biotic agents and to hold chemical reactions at their lowest levels. Forest reproductive materials provide a wide range of applications to control "active" hydrous status using a drying technique or by using a "passive" hydrous status through the measurement of a... Controlling hydrous status allows for the conservation of seed or pollen lots for operational, research, or genotype conservation purposes.

Forest tree seed crops can be irregular. As a consequence, reforestation, genetic improvement, and genetic resources seed bank programs must operate within available perennial harvests. Resorting to conservation procedures is therefore unavoidable and is critical to the success of a viable seed bank program (Probert 2003). Orthodox seed and pollen conservation processes involve multiple stages (initial open-air drying, post-maturation, extraction, and final drying) during which the hydrous status of seeds and pollen should be closely monitored. Seed and pollen are conventionally dried using heat. This drying process must be properly controlled to avoid excessive drying likely to lower the short- and long-term quality of lots. Dried materials are not preserved in a stable and definitive state. The conservation of dried materials is a rather slow and imperceptible, yet dynamic, process during which the permeability of storage containers, the environmental conditions of conservation, or the intrinsic conditions of the lots can change the initial hydrous status into values involving a potential risk of degradation (Colas and others 2012). Therefore, the medium- and long-term management of seed and pollen conservation requires controlling and adjusting the hydrous status of stored lots on a regular basis.

Irstea developed a seed/pollen dryer that is regulated by the a_w parameter that allows perfect coherence between the management and final control of hydrous status (Baldet 2006). This simple, reliable, fast, and reproducible integrated drying and control method can also be a component of an overall quality assurance project. Following the transfer of this technology as part of a scientific and technical collaborative agreement

signed between Irstea and the MRN, the DRF built and implemented several seed/pollen dryers using the information and plans provided by Irstea. These dryers have become invaluable tools in the routine daily management of seed and pollen lots in Quebec (Colas and Bettez 2012).

The purpose of this article is to present the general working principle and operating mode of the seed/pollen dryer and the functions of its different components, so that operational units that process seeds or pollen can build the dryer locally and can easily operate and maintain it.

Two-Pressure Principle

The a_w -regulated seed/pollen dryer works under the fundamental two-pressure principle developed originally by the National Institute of Standards and Technology (Hasegawa and Little 1977). This method is used as a standard relative humidity generator by saturating air with humidity under given controlled pressure and temperature and, as a second step, expanding the saturated air at the required operating pressure (figure 2). The resulting ERH (or a_w) equals the ratio of the two monitored total pressures (Equation 1). Post-expansion pressure ("p") is not necessarily the value of atmospheric pressure, but an additional parameter used to adjust the final ERH.

ERH (
$$a_w$$
) = $\frac{p}{p_1}$

- p = Post-expansion humidified air pressure.
- p_1 = Total pressure during the stage when air is saturated with humidity

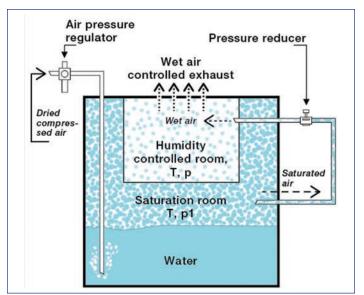


Figure 2. Schematic diagram of a two-pressure reactor. T = temperature, p1 = saturation pressure, and p = post-expansion air pressure. (Adapted from HUMOR 20 High-precision Humidity Calibrator data sheet, E + E Elektronik Ges.m.b.H, Langwiesen 7, A-4209, Engerwitzdorf, Austria)

Based on this principle, Irstea designed an a_w -regulated dryer. The diagram in figure 3 shows a simple version of the device with a set, manually preadjusted saturation pressure. An "automated" version of the dryer adjusts the saturation pressure automatically in regard to the effective ERH measured on an ongoing basis inside the drying cabinet (Baldet 2006). Only the simple version of the dryer operated at a set saturation

pressure is discussed in this article. The authors wish for this technique to generate accurate results at a satisfactory level, while remaining technologically simple and cost-effective to simplify the construction and routine management of this device. Figure 4 shows the device built at the DRF using the information provided by Irstea.

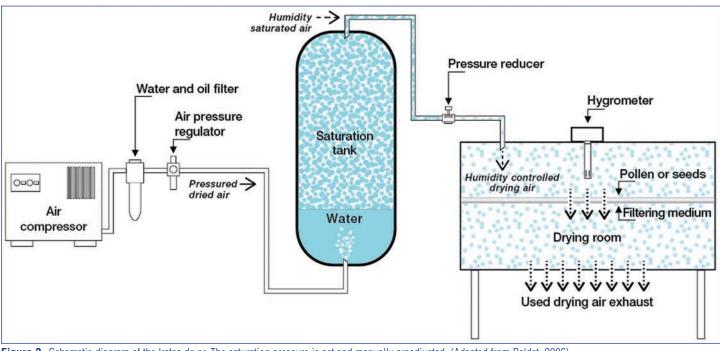


Figure 3. Schematic diagram of the Irstea dryer. The saturation pressure is set and manually preadjusted. (Adapted from Baldet, 2006)

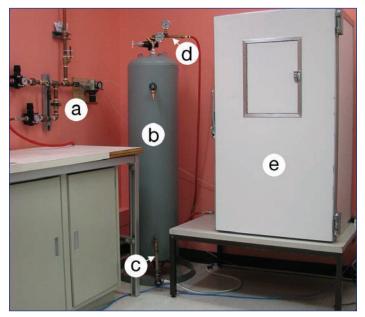


Figure 4. A general view of the device installed at the laboratory of the Direction de la recherche forestière. (a) Compressed air supply (in this case, supplied by the building system); (b) saturation tank (air humidification); (c) control tube showing the level of water in the saturation tank; (d) outlet for driving air at a predetermined level of a_w into the drying cabinet; (e) drying cabinet. (Photo by Fabienne Colas, 2012)

Design and Operation Details

Different technical stages were involved in the design of the seed/pollen dryer regulated by a_w (or at a controlled ERH). A detailed description of each stage follows. The characteristics of the main components needed to build the dryer can be provided on request. (Please contact Patrick Baldet.)

Saturation Tank

The tank used for air saturation ideally has an inner wall protected with a coating (e.g., enamel, paint) to prevent corrosion because of the presence of water and the constant renewal of dissolved oxygen. Water used in the tank must be at room temperature before the dryer is operated. It is therefore recommended to fill the tank at least 1 day before operations to stabilize the water temperature to that of the room where the dryer is installed. The water level in the tank can be easily monitored through the installed control tube (figure 4). The amount of water required depends on the expected time of use of the dryer. In the conditions discussed in this article (tank of about 32 gal or 120 L), the dryer was operated at an a_w of about 0.35 during one uninterrupted month with an initial water volume of about 5 gal (about 20 L).

The standard operation of the unit is also likely to increase the amount of water in the saturation tank if the ERH of the air expelled into the drying cabinet is below the ERH of the air pumped by the compressor. In our case, the output ERH was 35 percent (resulting a_w of 0.35) for an average ambient ERH of 50 percent.

Saturation Pressure Adjustments

Air saturation pressure inside the tank must be optimized for each installation. The pressure will vary slightly depending on tank characteristics, the accuracy of saturation pressure measuring devices, and compressed air supply.

To optimize air saturation pressure in the specific conditions of each laboratory, it is recommended to develop a curve relating the a_w generated by the expansion of pressurized air to the saturation pressure in the tank (figure 5). The first expansion pressure at the tank outlet is set at 5 psi (0.34 bar), which represents a 13 gal/min (or 50 L/min) air flow for 0.25 in (6 mm) piping not exceeding 6 ft (2 m) in length between the air expansion point and the drying cabinet. This average flow is recommended for a 5 ft² drying surface (about 0.5 m²), representing an average air-flow rate of 4 in (10 cm) per minute across the section of the drying cabinet. The required tank saturation pressure can be set and adjusted according to the desired a_w . A pressure of 18 to 34 psi (1.3 to 2.3 bar) is required to obtain a 0.4 to 0.3 a_w (figure 5).

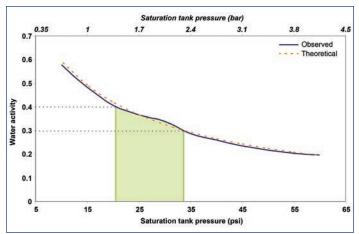


Figure 5. Water activity (a_w) of drying air, measured and theoretical, at the outlet of the dryer with relation to the saturation pressure in the tank (in psi or bar). The expansion pressure of air at the dryer outlet is set at 5.00 psi (0.34 bar). In green is the target range of pressures in the saturation tank necessary to obtain a level of a_w between 0.3 and 0.4 required to dry seeds or pollen. The theoretical a_w values were calculated using Equation 1. (Barbosa and others 2007)

The dryer described in this article was designed for safe, heatfree seed or pollen drying to limit damage sustained by materials that are still immature or that have only partly acquired physiological resistance to drying (Hay and Smith 2003). The wide diversity of forest reproductive materials increases the likelihood of having to manage in a single procedure lots containing seeds and pollen with different levels of tolerance to drying.

As shown in figure 5, the curves of both observed and theoretical values practically merge. In fact, they are identical if the mean accuracy of a_w measurement devices (±0.02) and type 1 medium accuracy-class manometers (±1 percent) are integrated. This illustration shows that the application of theoretical saturation pressure values is highly reliable. Measuring a_w systematically at the outlet of the drying cabinet is not necessary because only the accurate adjustment and measurement of the saturation pressure is sufficient. Therefore, the dryer can be operated without any complex electronic measuring instruments.

The Drying Cabinet

The material to use for the drying cabinet structure must not be hydrophilic, and it should be smooth and washable. Wood should be avoided. A good compromise is to use stiff plastic such as KömaCel[®] (Kömmerling Kunststoffe, Pirmasens, Germany), which interacts very little with the a_w of drying air and is easy to cut and assemble. Any other plastic material sufficiently stiff can be used. A laboratory oven can even be converted into a dryer after removing the electrical equipment and making minor modifications for the intake and outlet of drying air. The dimensions of the drying cabinet depend on the future use of the equipment (number of lots, volume of lot units, purity requirements, etc.).

Drying air will enter the cabinet at the base and will exit at the top, following the natural upward movement of moist air. The drying air is introduced inside the cabinet through pipes punctured at intervals of 2 to 4 in (5 to 10 cm), so that the drying airflow distributes evenly (figure 6). This method is well suited for drying small lots with high purity guaranteed by individual conditioning, allowing for dried materials to be in indirect contact with the main flow of drying air. To dry large amounts of material covering most of the useful surface of the dryer with no or little purity constraints, the direction of drying air supply can be more efficient if flowing from top to bottom (figure 3). Therefore, the drying air will directly flow through the layer of materials laid out to dry. This downward air supply mode is particularly well suited to light products like pollen or winged seeds that will be flattened against the drying surface.



Figure 6. Drying cabinet built at the Direction de la recherche forestière. (a) Drying air intake at controlled humidity levels, (b) air intake suitable for the final expansion of drying air, (c) containers used for drying pollen lots, (d) hygrometry probe in the drying cabinet, (e) outlets to evacuate drying air after use, (f) drying cabinet door. (Photo by Fabienne Colas, 2012)

In the drying cabinet, the position of the a_w measurement probe in relation to air intake is important. If the probe is placed relatively close to the air intake (lower part of the cabinet), it will measure the a_w of the drying air and will show the proper operation of the technical process. If the probe is placed opposite the air intake (position shown on figure 6, on the upper part of the cabinet), however, the probe will measure the a_w after exchanging with the materials during the drying process and will then represent the drying interactions of materials treated with higher values at the onset of the process, if a large quantity of moist materials is inside the cabinet. The position close to the air outlet is preferable because it enables to more accurately monitor the drying of materials.

Operations

After water in the tank is at room temperature, open the compressed air intake and adjust the tank saturation pressure

at the value determined during preliminary adjustments while keeping the outlet closed. The expansion pressure at the outlet of the tank will be adjusted after the required pressure is achieved in the tank. When the required pressure is achieved, open the air intake in the drying cabinet and verify the ERH value. If needed, adjust the saturation pressure to achieve the desired ERH value and, consequently, the resulting a_w required for the products to dry. After adjusted, the ERH remains very stable. Because most hygrometers provide measurements at a precision of ± 2 units of ERH, this measurement uncertainty should be considered in any new adjustment of the saturation pressure. Overall, the saturation pressure is always easier to measure than ERH. Using a hygrometers are only a means to control actual saturation pressure.

Drying

To optimize drying efficiency, samples, whether seeds or pollen, should be laid out in relatively thin layers in a container permeable to air. We use and recommend a polyamide monofilament textile mesh produced by the SAATI Company (Appiano Gentile, Italy).

When samples have different initial a_w values, the driest samples should be placed in the cabinet next to the drying air intake, and the dampest samples should be placed next to the air outlet to avoid rehydrating already stabilized samples.

The time needed to dry samples depends on their initial hydrous status. To ensure a final stable sample value, it is preferable to leave samples slightly longer in the dryer. After the a_w value imposed by ERH-controlled air is achieved, the samples will maintain this value even if they remain for a relatively long time in the dryer (figure 7).

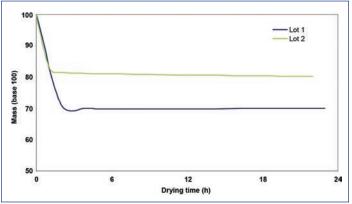


Figure 7. Stabilization of the mass of two Japanese larch pollen samples at a drying a_w level of 0.35. In such cases, the pollen is dry after about 2 to 4 hours in the dryer. Their mass, and therefore their a_w -qualified hydrous status, remains at a constant level for up to 24 hours in the dryer. (Philippe and others 2006)

Lot Storage

When removing the samples from the dryer, the lots must be quickly placed in vapor-proof storage containers to minimize water uptake. To make it easier to handle samples and to limit water uptake, the room where the dryer is set up must ideally have an ERH value close to the value applied to samples in the dryer.

Operational Advantages

- Based on its underlying principle, the drying process discussed in this article aims at achieving equilibrium between the ERH of drying air and the a_w of treated materials. With this technique, seeds or pollen can remain inside the drying cabinet without any risk of drying beyond the required value, because the equilibrium value remains constant after it is reached. This is a significant advantage when treating samples during an intensive period of activity often associated with climatic and phenological constraints. In fact, this technique allows easier management of technical resources assigned to a drying operation, particularly for pollen. Therefore, there is no need to mobilize several people to treat samples quickly. Leaving a sample slightly longer in a dryer will not impact its quality, because this technique defines a lower technical threshold of a,, unlike that observed when samples are dried using the conventional method of heat application.
- The seed/pollen dryer stabilizes samples at a predetermined a_w necessary for conservation. As the samples are dried without the application of heat, achieving equilibrium is safer for the biological material and more progressive than conventional drying. Storage at low temperatures can be engaged quickly without a cooling stage highly conducive to untimely water uptake.
- Climate change issues are generating more interest in long-term conservation of seeds and pollen. The hydrous status of preserved lots can change because of container permeability, aging, etc., during conservation. Owing to the nondestructive advantage of measuring a_w, the quality of lots preserved in banks can be controlled on a regular basis without reducing initial stored amounts. Any significant increase in a_w will lead to the degradation of lot quality (figure 1). Therefore, lots with a raised a_w can be restabilized at the optimal a_w of conservation in the dryer, to be then placed back into the bank. This technique can help ensure the quality of seed and pollen lots over the long term thus enhancing the potential of regenerating lots for future reintroduction.

• This drying process is based on an accurate, yet basic method. Only a quality pressure regulator is needed to ensure a reliable and continuous relative humidity of the drying air. Because it is so technically simple to use, the dryer is particularly intended for point-to-point or seasonal applications during which operators' control, calibration, and training costs must be as minimal as possible.

Operational Limitations

- The production of compressed air is a low energy-efficiency operation because air compression is an exothermic process requiring a large amount of energy. Therefore, the drying process involving the production of air at a level of ERH controlled by the two-pressure technique should be restricted to small drying units and seasonal applications. In addition, as shown in figure 5, obtaining the lowest ERH values requires increasingly higher pressures. Because this process is not a linear function, but a kind of asymptotic function in which the production of very dry air at 1 percent ERH would theoretically require a saturation pressure of 1,430 psi, it is appropriate to restrict the use of this method to relative humidity values above 20 percent (a_w values above 0.2).
- Dryer operations are based on the availability of filtered, de-oiled compressed air. For instance, operations may require dedicated air compression equipment if the "drying room" is not connected to a compressed air production and distribution system.
- Under national legislation, very strict regulations dictate the use of pressure equipment. In particular, regulations pertaining to the exclusive use of certified and tested materials are also subject to periodic official controls. These regulatory constraints must be considered at the early stages of any a_w pollen and seed dryer project and particularly when choosing the equipment to implement.

Address correspondence to:

Patrick Baldet, Research Engineer, Irstea, Unité Écosystèmes forestiers, Domaine des Barres, 45290 Nogent sur Vernisson, France; e-mail: patrick.baldet@irstea.fr.

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