1974

Bonner, F. T. (1974), *Seed Sci. & Technol.*, **2**, 399–405

Determining seed moisture in *Quercus*

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Summary

The air-oven method with drying times 7 to 8 hours shorter than those now prescribed in the ISTA rules proved adequate for determining moisture contents in acorns of several North American oaks. Schedules of 8 hours at 105 °C for *Quercus muehlenbergii* and 9 hours at 105 °C for *Q. shumardii* and *Q. nigra* gave moisture contents within three percentage points of those obtained by toluene distillation. Eight hours at 105 °C was accurate only to within six percentage points for *Q. alba*, apparently because of extreme variation in moisture among individual acorns of this species. The most desirable sample size was found to be five acorns. Acorns were prepared for drying by cutting into halves or quarters. Measurements showed that from 25 to 40 % of acorn moisture was in the pericarp tissue.

Résumé

Determination de la teneur en eau des semences de Quercus

La méthode en étuve avec un temps de sèchage plus court de 7 à 8 heures que ceux actuellement prescrits dans les règles de l'ISTA s'est avérée adéquate pour déterminer les teneurs en eau des glands de plusieurs espèces de chêne d'Amérique du Nord. Des durées de 8 heures à 105 °C pour *Quercus muehlenbergii* et 9 heures à 105 °C pour *Q. shumardii* et *Q. nigra* ont fourni des chiffres de teneur en eau différant de moins de 3 points de pourcentage de ceux obtenus par la méthode de distillation au toluène. Huit heures à 105 °C n'a donné des résultats exacts qu'à six points près pour *Q. alba*, apparemment en raison de l'extrême variabilité individuelle de la teneur en eau des glands de cette espèce. La taille de l'échantillon déterminée comme la plus souhaitable est de cinq glands. Les glands ont été préparés en vue du sèchage en les coupant en deux ou en quatre. Les mesures ont montré que 25 à 40 % de l'eau du gland était localisée dans le tissu du péricarpe.

Zusammenfassung

Die Bestimmung der Feuchtigkeit der Samen von Quercus

Die Trockenschrankmethode mit Trocknungszeiten, die 7 bis 8 Stunden kürzer sind als diejenigen, die jetzt in den ISTA-Vorschriften vorgeschrieben sind, erwies sich als geeignet für die Bestimmung des Feuchtigkeitsgehalts von Eicheln mehrerer nordamerikanischer Eichenarten. Verfahren von 8 Stunden bei 105 °C für *Quercus muehlenbergii* und 9 Stunden bei 105 °C für *Q. shumardii* und *Q. nigra* lieferten Feuchtigkeitsgehalte innerhalb 3 % von denjenigen Werten, welche durch die Toluol-Destillations-Methode erzielt wurden. Acht Stunden bei 105 °C war genau nur bis zu innerhalb 6 % für *Q. alba*, offensichtlich infolge extremer Streuung der Feuchtigkeit bei den einzelnen Eicheln dieser Art. Es zeigte sich, dass die empfehlenswerteste Probengrösse bei 5 Eicheln lag. Die Eicheln wurden für das Trocknen durch Halbieren oder durch das Schneiden in Vierteln vorbereitet. Die Messungen zeigten, dass 25 bis 40 % der Eichelfeuchtigkeit sich im Pericarpgewebe befand.

Introduction

Current ISTA rules (International Seed Testing Association, 1966) specify the air-oven method at 105 °C for determining seed moisture in *Quercus*. Coarse grinding is required because the seeds are large, and two-stage drying is necessary because moisture contents are normally well above 18 %. The present study was an attempt to find an easier method of sample preparation and moisture determination for several North American species of *Quercus*.

Earlier studies at the Forest Tree Seed Laboratory had revealed several problems in assessing moisture content of acorns. Two-stage drying is time-consuming and offers abundant opportunity for human error. Moisture contents of individual acorns from the same storage container can vary greatly. Also, some species have crude fat contents of 15 to 20 % (Bonner, 1971), and it was thought that toluene distillation might be advantageous.

Material and methods

General

Acorns studied were of four species: *Quercus alba* L. and *Quercus muehlenbergii* Engelm. from the subgenus *Luecobalanus*, *Quercus shumardii* Buckl. and *Quercus nigra* L. from the subgenus *Erythrobalanus*. One species of each subgenus has large acorns (*Q. alba*, *Q. shumardii*), and one from each has small acorns (*Q. muehlenbergii*, *Q. nigra*). All work was done with acorns collected from 1970 to 1972 in central Mississippi.

Distribution of moisture in acorns

One possible reason for the extreme variation in moisture among acorns is the thick pericarp tissue. To determine the distribution of moisture within individual acorns, pericarps of 20 acorns of each species were carefully removed. Both pericarp and embryo were dried for 15 hours at 105 °C.

Desirable sample size

The effect of individual acorn variation can be reduced by increasing the number of acorns in the sample, but the capacities of drying dishes and distillation equipment limit sample size. To determine a desirable size, 25 composited samples of 1, 2, 3, 5 and 10 acorns each were processed for each species. The acorns were cut in pieces and dried for 15 hours at 105 °C. The coefficient of variation was calculated for each composited sample size.

Sample preparation

Two tests checked the effects of sample preparation and drying method on moisture measurement. In the first, fifty 10-acorn samples of *Q. shumardii* were randomly drawn. Half were dried intact for 15 hours at 105 °C, and half were processed by

two-stage drying. The second test included 60 samples each of *Q. nigra*, *Q. alba*, and *Q. muehlenbergii*; there were 5 to 10 acorns per sample. Acorns were prepared by three methods: intact, two-stage drying, and cutting in halves (small acorns) or quarters (large acorns) with hand shears. With the exception of the 130 °C drying in the two-stage method, all ovendrying was for 15 hours at 105 °C. Satisfactory grinding was achieved in 15 seconds per sample with a high-speed blender.

Selection of air-oven schedules

The procedures used were those devised by Hart and Golumbic (1966) and modified by Bonner (1972). Each species lot was thoroughly mixed and divided into 25 sublots. Twelve of the sublots were dried to varying degrees by exposure for up to 48 hours in an air-conditioned room. Twelve more were placed in a humid cabinet at room temperature to absorb additional moisture. One sublot was not treated further. All sublots were then stored in polyethylene bags for about 20 days.

For each species, 14 samples were drawn from each of four sublots of median moisture content. Samples were weighed to the nearest 0.1 g and dried in glass dishes in forced-draft laboratory ovens at 105° or 130 °C. After 1, 2, 4, 6, 8, 12 or 24 hours of drying, they were removed from the ovens in pairs and reweighed. Cooling in desiccators was unnecessary because preliminary tests showed that in the air-conditioned laboratory hot samples would not gain weight until four minutes had elapsed. Weighing a pair of samples took about 30 seconds. Loss of moisture over time was calculated as a percentage of fresh weight, and the four-lot average loss at both temperatures was plotted against drying time.

Moisture contents of these same sublots were determined by toluene distillation of five-acorn samples (Horwitz, 1965). Distillation was into 10 ml Dean and Stark receivers for six hours. Preliminary trials showed that four to six hours of distillation was required to remove all moisture. A horizontal line corresponding to the mean moisture content as determined by toluene distillation was added to the time-temperature curves. The vicinity of intersection of the toluene line with the time-temperature curves suggested promising air-oven schedules for additional testing.

Moisture contents of the remaining 21 sublots of each species were then determined by toluene distillation and by the potential air-oven schedules. Chi-square tests were used to indicate the accuracy of the air-oven procedures to within one, two or three percentage points of the toluene values at 95% probability (Freese, 1960).

Results

Distribution of moisture in acorns

In *Q. shumardii*, *Q. nigra* and *Q. muehlenbergii*, the pericarp contained 25 to 33% of all acorn moisture (table 1). With *Q. alba*, 41.5% of total moisture was in the pericarp. *Q. alba* pericarps were noticeably thicker and comprised more of the total acorn weight than those of the other three species.

Table 1. Moisture distribution between pericarp and embryo in 20 acorns from each of four species.

Character	Species			
	Q. shumardii %	*Q. nigra* %	*Q. alba* %	*Q. muehlenbergii* %
Moisture content of entire acorn	35.5	30.3	52.5	41.8
Moisture content of embryo	37.2	31.3	51.0	42.8
Moisture content of pericarp	32.3	28.0	54.8	39.7
Proportion of total acorn weight in embryo	69.0	69.4	60.2	67.5
Proportion of total acorn weight in pericarp	31.0	30.6	39.8	32.5
Proportion of total acorn moisture in embryo	72.3	71.7	58.5	69.1
Proportion of total acorn moisture in pericarp	27.7	28.3	41.5	30.9

Desirable sample size

Comparison of the coefficients of variation for different sizes of composited samples indicated little advantage in having more than five acorns per sample (figure 1). Therefore, five-acorn samples were used in the remainder of this study.

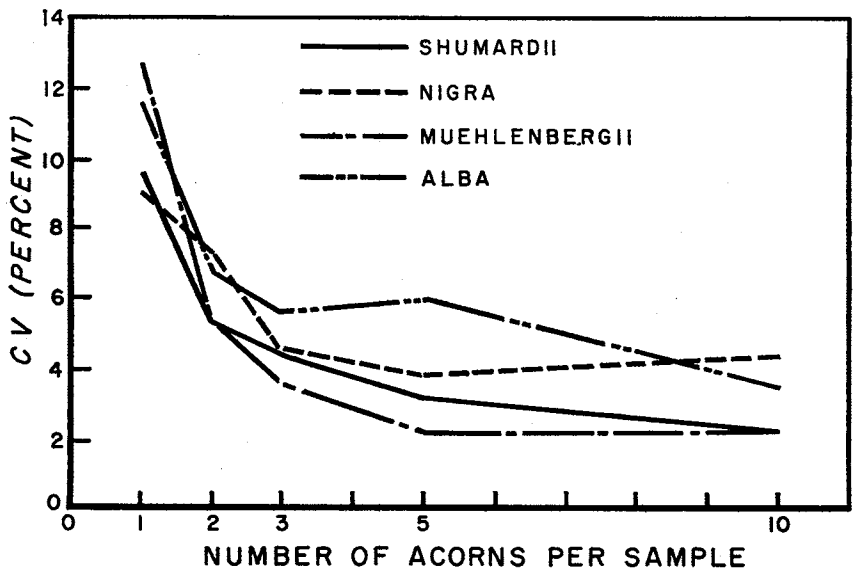

Figure 1. Effect of sample size on coefficient of variation of the mean (CV) in moisture content of four species of *Quercus*. Data are based on 25 trials with composited samples of each size for each species.

Sample preparation

In the first test with *Q. shumardii*, intact acorns averaged 35.8% moisture, and those measured by two-stage drying averaged 36.6%; the difference was significant at the 0.05 level. In the second test there were no significant differences between the mean moisture contents of *Q. alba* and *Q. muehlenbergii* acorns prepared by three different methods (table 2). With *Q. nigra*, intact acorns had significantly lower mean values than those processed by two-stage drying or cutting; but there was no significant difference between cutting in pieces and two-stage drying. Since cutting was much quicker and simpler than two-stage drying, all subsequent samples were cut.

Table 2. Mean moisture contents as determined by three different methods.

Species	Method		
	Intact %	Two-stage %	Cut in pieces %
Q. alba	47.4	48.0	47.9
Q. muehlenbergii	36.8	37.1	37.3
Q. nigra	31.5*	32.2	32.4

* Significantly different at the 0.05 level from the other two methods with *Q. nigra*.

Selection of air-oven schedules

The moisture-loss curves for *Q. alba* and *Q. shumardii* (figure 2) illustrate how potential air-oven schedules were chosen. Theoretically, the point where the toluene line intersects the curves should indicate the best drying time; but this point may fluctuate among seed lots. The best drying times are those chosen from areas of the curves where the toluene line is identical to or at least parallel to the moisture-loss curve. On this basis, the following air-oven schedules were chosen for testing with all sublots:

Q. alba – 7 to 9 hours at 105 °C
Q. muehlenbergii – 6 to 8 hours at 105 °C
Q. shumardii – 8 to 12 hours at 105 °C
Q. nigra – 8 to 9 hours at 105 °C

All 130 °C drying times were rejected because of the angles of intersection of the moisture-loss curves with the lines defining the toluene values.

After several trials within the times suggested above, the final toluene and air-oven comparisons were made with all 25 sublots. *Q. alba* and *Q. muehlenbergii* acorns were dried for eight hours at 105 °C, and *Q. shumardii* and *Q. nigra* were dried for nine hours at 105 °C. These air-oven schedules proved accurate to within three percentage points of the toluene values for *Q. shumardii*, *Q. nigra*, and *Q. muehlenbergii* (table 3). *Q. alba* was much more variable; the chi-square analysis suggested that eight hours drying at 105 °C was accurate only to within six percentage points of the toluene value for that species.

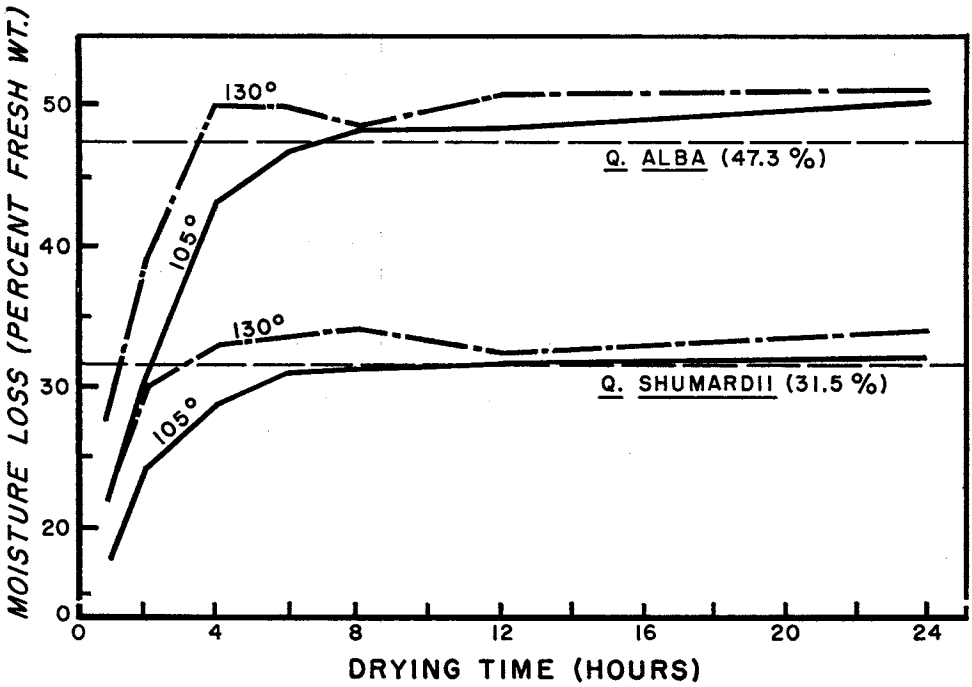

Figure 2. Moisture loss over 24 hr drying at two temperatures for *Q. alba* and *Q. shumardii*. Each value is a mean of four determinations. Toluene values are given by the dashed lines.

Table 3. Summary of air-oven and toluene measurements.

Species	Samples	Range of sublot moisture content %	Mean moisture content		Accuracy range with toluene values at 0.05 level of significance %
	Number		Toluene %	Air-oven %	
Q. alba	24	44.0–55.2	52.3	48.5	6
Q. muehlenbergii	25	32.3–41.7	37.3	36.5	3
Q. shumardii	25	27.1–37.1	32.1	31.9	3
Q. nigra	25	23.4–33.6	28.9	28.8	3

Discussion

Intact acorns did not completely lose their moisture unless the pericarps split open to expose the cotyledons to direct heat or contact with the toluene. The two white oaks, *Q. alba* and *Q. muehlenbergii* had many pericarps already split open by emerging radicles; these non-dormant acorns frequently sprout in storage. Thus, there was no

difference between intact and cut acorns for these species (table 2). The other two species normally exhibit dormancy and had not started sprouting; some form of cutting or grinding was necessary to extract all moisture from their acorns. Cutting into halves or quarters is certainly easier than two-stage drying, and the procedure should be considered for all *Quercus* work.

In experimental work, samples must be small enough to fit the equipment yet large enough to adequately represent the population. Sample sizes based on weight are normally preferable to samples based on the number of seed; but with *Quercus* and a few other large tree seeds, this number/weight relationship must be reconsidered. The wet weight of five *Q. shumardii* acorns can easily be 25 g or more. A reasonable sample size would be 10 g for small acorns, such as *Q. nigra* and *Q. muehlenbergii*, and 25 g for large acorns, such as *Q. shumardii* and *Q. alba*.

The great variation in moisture content among individual acorns seems to be inherent with this genus. Pockets of insect and disease damage, thick spongy pericarps (on some species) and premature sprouting in storage may all contribute to this variation. This variation makes it practically impossible to meet the ISTA requirement of duplicate determinations not differing by more than 0.2% (ISTA, 1966).

The air-oven method with drying times seven to eight hours shorter than those now prescribed in the ISTA rules proved adequate for determining moisture contents in acorns of several North American oaks. Drying periods of eight and nine hours may be inconvenient, however, if the normal workday is eight hours long. Better accuracy with these methods was desired, but values to within 3% at 95% probability are sufficient for most seed handling decisions. Additional work should be done with *Q. alba* to try to improve the methods for that species.

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