



Canadian Grain Commission
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Project Report Executive Summary

Prediction of germination energy of malting barley during long-time storage

By

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This is the final report for a two-year project conducted between September 1, 2002 and October 31, 2004.

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1. Evaluation of various instrumental techniques for detection and measuring the degree of pre-germination in barley

In the absence of dormancy, Canadian malting barley varieties have a propensity to pre-germinate in the field under extended moist conditions during harvest. Although a visual inspection of barley, conducted during the selection processes, can identify severely sprouted grain, it cannot detect the incipient germination. The germination energy test does not indicate pre-germination in barley. Barley selectors and evaluators do not have a practical tool to predict how much of the initially selected barley will lose germination energy during storage. This makes it difficult to know how much grain to select initially. It is also disappointing and costly to farmers who are initially advised that their barley was selected for malting purposes and informed later that the grain is no longer acceptable for malting. There is, therefore, a clear need for a rapid and practical method for detection of pre-germination and prediction of safe storage time for malting barley.

This study investigated the following techniques to determine their suitability to detect and measure the degree of pre-germination in Canadian malting barley: rapid visco analysis (RVA), image analysis, near infrared reflectance (NIR) and Fourier transform infrared spectroscopy (FTIR).

Based on the results of this study, it appears that at the present time, neither NIR nor FTIR spectroscopy techniques are able to reliably detect and measure the degree of pre-germination in barley grain.

The results of this study suggest that grain appearance, as assessed by visual examination and/or image analysis, is not related to its composition and/or to the biochemical processes occurring in the grain. Therefore, the assessment of appearance via either method cannot, at the present time and/or the present state of technology, reliably detect and measure the degree of pre-germination in barley grain.

This study provided evidence that RVA is the suitable technique to detect pre-germination in barley. Since it is known that one of the signs of pre-germination is an elevated level of alpha-amylase in the kernel, and since the RVA parameters are very sensitive to the increasing level of alpha-amylase in the samples, it was concluded that:

- The RVA test is able to detect the signs of pre-germination in barley samples.
- The RVA test is able to measure the degree of pre-germination in barley as related to the amount of alpha-amylase in the samples.

The measurements of germination energy (GE) after accelerated aging of barley samples confirmed that:

- Barley samples with the initially high RVA final viscosity (FV) values are capable of retaining their high germination energy ($GE \geq 95\%$) even when exposed to poor storage conditions.

- Barley samples with the initially low RVA FV values show propensity to lose viability and exhibit low GE (<95%) after exposure and/or storage at poor conditions.

2. General interpretation of RVA results

The following general interpretation of the RVA results and recommendations were developed and can be used to identify barley samples having potential problems during long-time storage and/or storage in hot and humid conditions.

RVA-final viscosity < 100 RVA units

- Samples with final viscosity values lower than 100 RVA units are pre-germinated, and the probability that they will lose GE after storage is ~95%. They should be malted as soon as possible.
- ~5% of samples which had the RVA FV values lower than 100 RVA units and retained GE after storage were samples with a very low moisture content.

RVA-final viscosity between 100 and 135 RVA units

- Samples with final viscosity values between 100 and 135 RVA units are more difficult to categorize. The probability that they will lose GE after storage is ~75% -- the loss will occur especially in samples with high moisture contents.
- ~25% of samples which had RVA values between 100 and 135 RVA units and retained GE after storage were samples with a very low moisture content.
- Samples with RVA value between 100 and 135 RVA units have a better chance to retain GE during storage if the storage conditions are good (cool and dry).

RVA-final viscosity > 135 RVA units

- Samples with final viscosity values higher than 135 RVA units are sound and they should retain GE even after a long-term storage. The probability that they will retain GE after storage is about 99%.
- 1% of samples that had RVA value > 135 RVA units and lost GE after storage were samples with a very high moisture content.

3. Recommended practices for testing barley samples for pre-germination using RVA-StarchMaster unit

The following practices are recommended for testing barley samples for pre-germination using RVA-StarchMaster unit:

A) Sample preparation

1. Clean barley grain using Carter dockage tester. The setting of the kicker is barley No. 6 riddle, No. 6 buckwheat dockage screen at top sieve, solid plain sieve at middle sieve, air control maximum, and speed control # 7. Samples should be stabilized to the room temperature before any measurements and grinding.
2. Determine the moisture content (%) in samples using NIR.
3. Mix the cleaned sample thoroughly and take a small representative sample (recommended amount ~300g). This is an important step to ensure that the sample being analyzed represents the whole material. A precision divider or similar device should be used to take an appropriate sub-sample.
4. Grind the sample using the Falling Number Laboratory Mill 311 (0.8 mm screen). Whole grain samples should be brought to room temperature before grinding. Clean grinder thoroughly after each sample is ground. If not using immediately, store ground sample in cold room.
5. Thoroughly stir the ground samples before sub-sampling and weighing. Weigh appropriate amount of the ground sample into a canister. Use the moisture content as determined by NIR and the “moisture conversion table” to determine the amount of sample (g) to be weighed into a canister.

Moisture Conversion Table

M.C. (%)	Weight (g)	M.C. (%)	Weight (g)	M.C. (%)	Weight (g)	M.C. (%)	Weight (g)	M.C. (%)	Weight (g)	M.C. (%)	Weight (g)
8.0	3.74	9.5	3.80	11.0	3.87	12.5	3.93	14.0	4.00	15.5	4.07
8.1	3.74	9.6	3.81	11.1	3.87	12.6	3.94	14.1	4.00	15.6	4.08
8.2	3.75	9.7	3.81	11.2	3.87	12.7	3.94	14.2	4.01	15.7	4.08
8.3	3.75	9.8	3.81	11.3	3.88	12.8	3.94	14.3	4.01	15.8	4.09
8.4	3.76	9.9	3.82	11.4	3.88	12.9	3.95	14.4	4.02	15.9	4.09
8.5	3.76	10.0	3.82	11.5	3.89	13.0	3.95	14.5	4.02	16.0	4.10
8.6	3.76	10.1	3.83	11.6	3.89	13.1	3.96	14.6	4.03	16.1	4.10
8.7	3.77	10.2	3.83	11.7	3.90	13.2	3.96	14.7	4.03	16.2	4.11
8.8	3.77	10.3	3.84	11.8	3.90	13.3	3.97	14.8	4.04	16.3	4.11
8.9	3.78	10.4	3.84	11.9	3.90	13.4	3.97	14.9	4.04	16.4	4.11
9.0	3.78	10.5	3.84	12.0	3.91	13.5	3.98	15.0	4.05	16.5	4.12
9.1	3.78	10.6	3.85	12.1	3.91	13.6	3.98	15.1	4.05	16.6	4.12
9.2	3.79	10.7	3.85	12.2	3.92	13.7	3.99	15.2	4.06	16.7	4.13
9.3	3.79	10.8	3.86	12.3	3.92	13.8	3.99	15.3	4.06	16.8	4.13
9.4	3.80	10.9	3.86	12.4	3.93	13.9	4.00	15.4	4.07	16.9	4.14

6. Add exactly 25.0 mL of deionized water using an “eppendorf pipette” or a “dispensette pipette”.

7. Mix the content of the canister with a spatula to disperse the sample. Use the paddle to scrape off the sample from a spatula. Insert the paddle into the canister.

B) Analysis

8. Analyze the samples using the RVA-StarchMaster unit and the program for RVA Stirring Number (SN). This is a standard 3 min program - it heats the sample to 95°C and mixes it for 10 sec at 960 rpm and then at 160 rpm. Record the final viscosity. The results will be displayed in cP (centipoise) units. To convert the numbers to RVA units, divide the cP units by 12.

RVA units = cP ÷ 12

Run the sample in duplicate.

C) Instrument maintenance

9. In order to ensure the accuracy of measurements, you should check the calibration offset daily. Please follow the instructions in your Installation and Operation Manual.

10. In addition, your StarchMaster unit should be calibrated with a standard oil. This should be done once per month. Newport Scientific recommends Cannon Certified Viscosity Standard R47000 (95C). A calibration Checking Kit is available from your Newport Scientific representative. Measure the viscosity of the calibration oil using the CAL95 program (program designed for calibration). The viscosity of the calibration oil should be within $\pm 5\%$ of the recommended value.

For example, if the recommended value is 1223cP and the reading is between 1162 cP and 1284 cP, your unit is ready for analysis. If the reading is outside of this range, your RVA-StarchMaster requires recalibration, please contact your representative. Only the representative can do it for you!

4. Recommended safe storage time for barley samples with various degrees of pre-germination

In order to more accurately predict the storage life of samples with various degrees of pre-germination, during the second year of this investigation long -term storage studies were also conducted. Barley samples with various degrees of pre-germination, as determined by the RVA test, were stored in four different environments:

1. Temperature $\geq 25^{\circ}\text{C}$; relative humidity $\geq 60\%$
2. Temperature $20\text{--}23^{\circ}\text{C}$; relative humidity $\leq 40\%$
3. Temperature $\sim 5^{\circ}\text{C}$; relative humidity 50-60%
4. Outdoor, variable conditions: samples were placed in the outdoor conditions in January 2004 and stored until January 2005.

The safe storage life of samples (retention of GE $\geq 95\%$) was dependent on the following factors:

- The degree of pre-germination in samples
- Storage conditions: temperature and relative humidity
- The initial moisture content of barley samples

In order to predict the storage life of barley samples more accurately, all factors have to be considered. Based on the storage studies conducted during the second year of this investigation, the following safe storage time is recommended for barley samples with various degrees of pre-germination:

Storage conditions	Safe storage time			
	RVA FV 0 – 50 (RVU ¹)	RVA FV 50 – 90 (RVU ¹)	RVA FV 90 – 135 (RVU ¹)	RVA FV ≥ 135 (RVU ¹)
T ≥ 25 °C RH² ≥ 60%	1 – 4 weeks	1 – 8 weeks	4 – 24 weeks	16 – 41 (+) weeks (+) samples tested for the last time at 41 weeks exhibited GE≥95
T 20 - 23 °C RH² ≤ 40%	2 – 24 weeks	32 – 42 (+) weeks	42 (+) – 50 (+) weeks	42 (+) – 50 (+) weeks
T ~ 5 °C RH² 50 - 60%	8 – 42 weeks	32 – 42 (+) weeks	> 42 (+) weeks	41 (+) – 50 (+) weeks
Outdoor storage Starting date: Jan 2004	Samples lost GE in May-June 2004	Samples lost GE in June - August 2004	Samples lost GE in June-November 2004. However, one sample with a low moisture content retained GE until January 2005	Samples were tested for the last time in November 2004 - all samples still exhibited good GE.

¹ Rapid Visco Units

² Relative Humidity

Note: It is possible that a large volume of barley stored in any of the above conditions will retain/lose germination energy in a slightly different time because the temperature and/or moisture of grain inside a large bin will not change at the same rate as for a smaller sample.

5. Sample collection and analysis

Barley samples used in this study were collected over two consecutive years, 2002 and 2003. All samples were selected for malting and exhibited germination energy $\geq 95\%$. In 2002, 306 samples were collected. Among the 2-row varieties, the greatest number of samples was collected for Metcalfe (107), followed by Harrington (32), Stratus (29), and Stein (23). Among the 6-row varieties, the greatest number of samples was collected for B1602 (56), followed by Robust (21). In 2003, 519 samples were collected. Among the 2-row varieties, the greatest number of samples was collected for Metcalfe (148), followed by Harrington (88), Stratus (65), Kendall (55), and Merit (18). Among the 6-row varieties, the greatest number of samples was collected for Robust (52), Excel (41), and Legacy (27).

Over the two-year period, barley samples were collected from the following crop districts of Western Canada:

Alberta: 2, 3, 4, 5, 7

Saskatchewan: 1B, 2A, 2B, 3AS, 4B, 5A, 5B, 6A, 6B, 7A, 7B, 8A, 8B, 9A

Manitoba: 1, 2, 3, 5, 6, 8

Significantly more samples were pre-germinated in 2002 than in 2003, which can be directly related to differences in the average precipitation during those two years. In general, the average precipitation in 2002 was above normal, whereas in 2003 it was below normal.

In 2002, the year with a very high average precipitation, Harrington exhibited a smaller percentage of pre-germinated samples (75%) than three other 2-row varieties tested in the study (91%). In 2003, the year with a very low average precipitation, Harrington again exhibited lower tendency for pre-germination (9%) than four other varieties tested in the study (16%). In both years, the 6-row varieties showed slightly lower percentage of pre-germinated samples (79% in 2002 and 12.5% in 2003) than the 2-row varieties (87% in 2002 and 15.5% in 2003). It has to be noted, however, that in both years a smaller number of 6-row samples (77 in 2002 and 120 in 2003) than 2-row samples (164 in 2002 and 374 in 2003) were tested.

These studies have shown that barley samples grown in the northern regions of Alberta (crop district 7), Saskatchewan (crop districts 5B, 8A, 9A), and Manitoba (crop district 3) exhibited a greater tendency for pre-germination than samples grown in other districts. The amount of precipitation could not fully explain these results, as in some cases the amount of actual precipitation was lower than the normal average for the respective districts or lower than in other districts. The later harvest dates (and/or higher harvest moisture) could be partially responsible for the observed results.