A Simple and Rapid Test for Drying Damage in Wheat

C. E. LUPANO and M. C. AÑÓN[1]

ABSTRACT

A simple test, based on the fact that the rate of CO_2 production is lower in heat-damaged seeds, allowed their rapid identification. CO_2 release was determined by measuring the color change of a pH indicator in the incubation medium. Highly significant correlation between this test and germination was found for the wheat variety utilized.

Cereal Chem. 65(1):49–51

Deterioration of wheat grain by drying procedures leads to important losses from decreased viability and industrial quality of the seeds (Ghaly et al 1973, Booth et al 1980, Schofield et al 1983, Wassermann et al 1983). The main assays for detecting deterioration are the germination and baking tests, both of which are time-consuming.

The objective of this work was to develop a procedure to allow quick detection of the damage induced by drying, without complicated techniques or equipment. The method is based on the fact that the rate of CO_2 production after imbibition is lower in heat-damaged seeds than in unheated seeds. Thus, there should be a correlation between the acidity produced in a given time in the incubation medium and the extent of deterioration of the seeds. It is therefore possible to measure the final acidity of the incubation medium, which results from the CO_2 produced by respiration of the seeds. The acidity is determined by measuring the change of color of a pH indicator.

MATERIALS AND METHODS

Wheat Samples

Seeds dried under different conditions. Wheat used was *Triticum aestivum* L. cultivar Marcos Juarez grown in Pergamino, Argentina, during 1982–1983. Seeds were moistened and dried as described in a previous paper (Lupano and Añón 1986). Moisture content was determined according to Giner and Calvelo (1987), and values obtained were corrected to the AOAC method (AOAC 1980). Table I shows the moisture content before and after drying, the air and seed temperature, and the drying time. Reference to the various moisture-temperature drying regimes throughout this paper corresponds to that given in Table I. Unprocessed seeds were used as controls.

Heated and nonheated seeds from other cultivars and crops. Seeds used were Marcos Juarez-INTA, 1983–1984 crop; Leones-INTA, 1983–1984 crop; Trigal 708, 1982–1983 crop; Saira-INTA, 1983–1984 crop; Buck patacón, 1983–1984 crop; and Buck napostá, 1983–1984 crop.

Heated seeds were all moistened and dried under the same conditions, according to the procedure mentioned previously (Lupano and Añón 1986).

Nonviable seeds. Seeds used were Marcos Juarez-INTA, 1982–1983 crop, which had been heated for 2 hr at 130° C.

Germination Assays

Germination assays were carried out according to the procedure described in a previous paper (Lupano and Añón 1986).

Decoloration Test

Incubation medium. The medium used was distilled water, adjusted to pH 10 with 1 *N* NaOH; pH was measured with a Merck pH universal indicator. Two drops of phenolphthalein solution were added (1% phenolphthalein in 96° C ethanol/distilled water, 60:40, v/v) to 50 ml of medium, and the color was made uniform by gentle stirring with a glass rod. This medium should be prepared just before using it, or else it should be kept in an air-tight bottle. In most of the assays, penicillin G and streptomycin sulfate were added to the medium, but identical results were obtained without them, showing that antibiotics might not be necessary.

Procedure. Assays were performed in 10-ml, 2.3-cm diameter, screw-cap plastic flasks. Samples of 29–31 seeds of approximately the same total weight were placed in each flask. Nonheated Marcos Juarez seeds were used as controls, and nonviable seeds or medium alone as blanks.

Then 4 ml of medium was added to each flask, care being taken to cover all seeds with liquid. The flasks were immediately capped and immersed in a bath at 30° C. The flasks were swirled gently and decoloration of the medium in the control flasks was watched for from 3 hr of incubation on. Flasks were removed from the bath at the time of decoloration of the control flask, and the media without the seeds were transferred to new flasks. Absorbances were read in a spectrophotometer at 550 nm. Spectrophotometric readings could be replaced by comparison with a color scale.

Treatment Before Incubation

In some assays, seeds were treated with 1% sodium dodecyl sulfate (SDS) or acetone for 3 min, with stirring, and rinsed three times with distilled water before performing the decoloration assay.

RESULTS AND DISCUSSION

Assays Performed with the Same Variety of Seeds Dried Under Different Conditions

Table II shows the percentage of germinated seeds after three days, the absorbance values obtained in two decoloration assays, and the mean percentages of decoloration and their standard deviations for the different drying conditions. These values were calculated by taking the incubation medium as blank, and by considering as 100% the value corresponding to the control.

An analysis of variance of the results was performed, considering as sources of variation the temperature of the drying air and the initial moisture content of the seeds. This analysis showed that significant differences can be detected by this assay, at a level of significance of 1%, among lots of seeds subjected to different thermal treatments ($F_{\text{temperature}} = 26.85$; $F_{\text{initial moisture content}} = 19.00$; $F_{0.01} = 8.02$), but an interaction at this level of significance was not observed between air temperature and initial moisture content ($F_{\text{interaction}} = 3.15$; $F_{0.01} = 6.42$).

To determine if this method allows detection of differences between lots of seeds dried under different conditions, an analysis of least significant differences was performed by comparing the means of populations. Results are shown in the last column of Table II. Drying conditions were divided into several groups, according to the values obtained by the test.

[1]Centro de Investigación y Desarrollo en Criotecnología de Alimentos (CIDCA), Facultad de Ciencias Exactas, UNLP. La Plata (1900), Provincia de Buenos Aires, Argentina.

TABLE I
Drying Conditions of the Seeds

Drying Condition[a]	Moisture Content Before Drying[b] (%)	Moisture Content After Drying[b] (%)	Drying Time (min)	Air Temperature (° C)	Final Seed Temperature (° C)
22-110	22.2	16.2	25	110	53
22-125	22.5	17.5	20	125	56
22-140	22.1	16.7	15	140	62
28-110	27.9	18.4	40	110	59
28-125	27.5	16.0	35	125	57
28-140	27.9	15.9	30	140	63
35-110	35.4	16.2	65	110	60
35-125	34.8	15.9	55	125	61
35-140	34.8	16.6	45	140	70

[a]Conditions were categorized by approximate initial moisture content (first number) and drying temperature (second number).
[b]Dry matter basis.

TABLE II
Percentages of Germination and Results of Decoloration Assays Performed with Seeds Dried Under Different Conditions

Drying Condition	Percentage of Germinated Seeds	Absorbance[a] Assay 1	Absorbance[a] Assay 2	Mean Percent of Decoloration[b]
Control	100	0.068	0.051	100
22-110	87	0.163	0.151	87.0 ± 1.4 a
22-125	75	0.196	0.187	82.5 ± 2.1 a
22-140	61	0.322	0.250	70.5 ± 2.1 ab
28-110	77	0.098	0.125	93.0 ± 4.2 a
28-125	20	0.387	0.372	58.0 ± 4.2 ac
28-140	2	0.493	0.683	29.5 ± 26.2 c
35-110	44	0.273	0.279	71.5 ± 4.9 ad
35-125	4	0.545	0.527	37.0 ± 5.7 bcd
35-140	0	0.623	0.650	24.0 ± 11.3 c
Blank	···	0.879	0.760	···

[a]The time of incubation was 3 hr 15 min.
[b]Percentages were calculated by substracting the blank to each absorbance value, and considering the control as 100%. Values followed by the same letter are not significantly different at the 1% level.

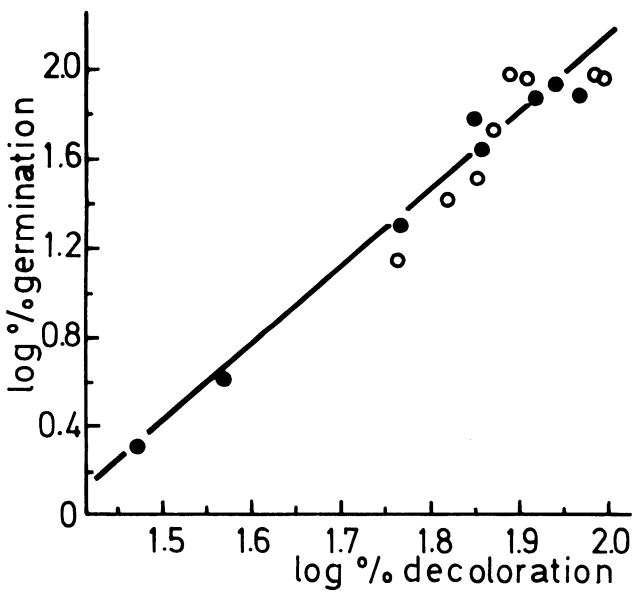

Fig. 1. Correlation between the germination at three days and the decoloration tests. ● = Seeds Marcos Juarez-INTA, crop 1982-1983, dried under different conditions. o = Seeds of different cultivars and crops.

Correlation with the germination assays. Figure 1 shows the straight line obtained by plotting germination (log %) versus decoloration (log %). Each black point is the mean of the two determinations mentioned above. The correlation coefficient obtained was 0.99, and the equation of the straight line was:

$$\log \% \text{ germination} = 3.4647 \log \% \text{ decoloration} - 4.7883 \quad (1)$$

The high correlation between the germination and decoloration tests should allow us to estimate the percentage of germination for a given variety of seeds by this assay. However, the model is inaccurate above 91% decoloration, because equation 1 gives values in percent germination higher than 100%. So, values of percent decoloration higher than 91 were considered as 100% germination.

Assays with Heated and Nonheated Seeds of Different Varieties

Table III shows the percentages of germination of each lot of seeds at three and six days and the percentage of decoloration obtained for each lot. It can be seen that, despite subjecting all heated seeds to the same moistening and drying treatment, some were more deteriorated with regard to their viability.

A new equation was calculated by combining all the data except those of Buck patacón. This equation includes the black and white points of Figure 1. The new correlation coefficient was 0.97, and the equation of the straight line was:

$$\log \% \text{ germination} = 3.4656 \log \% \text{ decoloration} - 4.8032 \quad (2)$$

Because this equation is almost equal to equation 1, equation 1 was used to estimate the percentages of germination. These results are shown in Table III. If the calculated values are compared with the actual ones for heated seeds, the percentages of germination estimated by the decoloration assay are similar to the actual percentages of germination for all varieties tested. For nonheated seeds, however, the percentages of germination estimated from decoloration coincided with the actual values for the Leones and Trigal 708 seeds but were much lower than the real germination percentages for the Saira-INTA, Buck patacón, and Buck napostá seeds.

To ascertain whether the problem was caused by differences in permeability, the seeds were treated with 1% SDS or acetone before performing the decoloration assay. Results are shown in Table IV.

It can be seen that Saira-INTA seeds treated with acetone or SDS behaved similarly to those of Marcos Juarez. This indicates that their abnormal response in the decoloration assay could result from a decreased permeability to water, which could be counterbalanced by pretreatment with acetone or SDS. Despite pretreatment with either SDS or acetone, a response similar to that of the other seeds could not be obtained for Buck patacón and Buck napostá seeds. The lower rate of CO_2 release into the medium exhibited by these seeds was probably caused by metabolic differences or by insufficient length of pretreatment with the SDS or acetone. The heated seeds did not present an abnormal response (Table III). This behavior could be explained if heat damage were the main cause of the lower rate of CO_2 release.

TABLE III
Percentages of Germination, Measured and Estimated by Means of Equation 1, and Percentages of Decoloration of Seeds of Different Varieties and Crops

Variety	Crop	% Germ. at Three Days		% Germ. at Six Days		% Decoloration[a,b]		% Germ. Calculated	
		Nonheated Seeds	Heated Seeds	Nonheated Seeds	Heated Seeds	Nonheated Seeds	Heated Seeds	Nonheated Seeds	Heated Seeds
Marcos Juarez-INTA	1983/84	97	53	97	53	100	74 ± 7	...	49
Leones-INTA	1983/84	96	33	97	39	96 ± 3	71 ± 13	100[c]	42
Trigal 708	1982/83	94	26	95	44	98 ± 3	66 ± 4	100[c]	33
Buck patacón	1983/84	96	3	97	11	41 ± 10	22 ± 10	6	1
Buck napostá	1983/84	97	0	97	0	77 ± 3	31 ± 2	56	2
Saira-INTA	1983/84	91	14	95	21	81 ± 5	58 ± 9	67	21

[a] Values in this table are the means of three determinations with their standard deviations.
[b] Calculations were made taking as 100% the value corresponding to Marcos Juarez-INTA, 1983–1984 crop, nonheated seeds, using as blank nonviable seeds.
[c] Values of % decoloration higher than 91 were considered as 100% germination.

TABLE IV
Decoloration Assay in Sodium Dodecyl Sulfate (SDS)- or Acetone-Treated Seeds

Treatment	Variety (1983-1984 Crop)	% Decoloration[a]	% Germination (Calculated)[b]
SDS	Marcos Juarez-INTA	100	...
	Saira-INTA	96	100[c]
	Buck patacón	50	13
	Buck napostá	81	67
Acetone	Marcos Juarez-INTA	100	...
	Saira-INTA	100	100[c]
	Buck patacón	52	14
	Buck napostá	79	61

[a] Assays were performed with nonheated seeds, using Marcos Juarez seeds as control, and nonviable seeds as blank.
[b] These values were estimated by means of equation 1. Values of % decoloration higher than 91 were considered as 100% germination.

The results obtained indicate the seeds that give percentages of decoloration higher than a given limit have not been damaged during drying. Seeds with percentages of decoloration lower than this limit would require further analysis to decide whether they have been damaged. Other variables affecting wheat quality were not investigated and would need to be studied before adopting the procedure.

LITERATURE CITED

ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTS. 1980. Page 211 in: Official Methods of Analysis. The Association: Washington, DC.

BOOTH, M. R., BOTTOMLEY, R. C., ELLIS, J. R. S., MALLOCH, G., SCHOFIELD, J. D., and TIMMS, M. F. 1980. The effect of heat on gluten-physico-chemical properties and baking quality. Ann. Technol. Agric. 29:399.

GHALY, T. F., EDWARDS. R. A., and RATCLIFFE, J. S. 1973. Heat-induced damage in wheat as a consequence of spouted bed drying. J. Agric. Eng. Res. 18:95.

GINER, S. A., and CALVELO, A. 1987. Modeling of wheat drying in fluidized beds. J. Food Sci. 52:1358.

LUPANO, C. E., and AÑON, M. C. 1986. Denaturation of wheat germ proteins during drying. Cereal Chem. 63:259.

SCHOFIELD, J. D., BOTTOMLEY, R. C., TIMMS, M. F., and BOOTH, M. R. 1983. The effect of heat on wheat gluten and the involvement of sulfhydryl-disulfide interchange reactions. J. Cereal Sci. 1:241.

WASSERMANN, L., MUHLBAUER, W., and SCHREIBER, H. 1983. Influence of drying on wheat quality. Part 3: Influence of moisture content of the kernels on drying and baking behavior. Getreide Mehl Brot 37:268.

[Received March 9, 1987. Revision received July 17, 1987. Accepted August 25, 1987.]