Influence of Germination on Wheat Quality. I. Functional (breadmaking) and Biochemical Properties[1]

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ABSTRACT

The effects of controlled laboratory germination (18, 35, and 54 h) on the biochemical, milling, rheological, and baking characteristics of an overly strong wheat, Glenlea, and a moderately strong wheat, Neepawa, were examined. A low level of germination produced a substantial improvement in the breadmaking potential of Glenlea; with further germination the quality decreased markedly. All germination treatments were detrimental to the breadmaking quality of Neepawa. Glenlea and Neepawa differed only marginally in their responses to germination in terms of milling characteristics, starch degradation, and enzyme activities. Progressive deterioration of gluten strength of both cultivars was indicated by concomitant changes in sedimentation values and farinograph parameters. The quality indices derived from the farinogram of the 18-h germinated Glenlea sample were essentially the same as those of the sound Neepawa sample.

Germination of wheat before harvest is a problem in North America. The overall detrimental effects of germination result from the cumulative losses of grain yield, grain quality (grade), flour yield, and flour quality. Research about such damage has been concerned primarily with the development of α-amylase activity and the consequences of the increased activity on the breadmaking process (Ibrahim and D'Appolonia 1979, Kozmin 1933).

Our main objective in the present study was to examine further the physical and biochemical changes in wheat and flour produced in the early stages of germination that may be related to the changes in breadmaking quality. A secondary objective follows from the observation that very strong mixing wheat cultivars perform well in baking tests in spite of having higher α-amylase activity than normal Canadian hard red spring cultivars. Accordingly, our second aim in this study was to investigate the effect of germination on a wheat cultivar with very strong dough-mixing characteristics.

MATERIALS AND METHODS

Material

Two cultivars of Canadian hard red spring wheat, Glenlea and Neepawa, were selected on the basis of their dough-making properties (very strong and strong, respectively). Neepawa is the predominant cultivar of the hard red spring class. Glenlea, because of its overly strong dough-mixing characteristics, was not acceptable for the red spring class but was licensed into the utility class because of its high yield potential (approximately 15–20% higher than Neepawa).

Germination Procedure

Samples of wheat (3 kg) were surface-sterilized by soaking in a solution of 2.0% aqueous sodium hypochlorite for 15 min at room temperature (about 20° C) and then rinsed well with distilled water for at least 20 min. Before germination, the wheat was soaked overnight (about 16 h) in excess distilled water at 4° C with one water change. After rinsing with distilled water, the steeped wheat was spread on wet cellulose pads and germinated at 21° C, 67% rh. At intervals of 18, 35, and 54 h, the samples were withdrawn and frozen at −30° C and then freeze-dried. Roots and coleoptiles were removed, and the freeze-dried grain was stored at −4° C. Sound (control) samples were surface-sterilized and freeze-dried immediately.

Milling

Wheat samples were tempered to 15.5% moisture content and milled into straight-grade flour on a Buhler pneumatic laboratory mill (MLV 202).

Analytical

Total nitrogen was determined by the macro- or micro-Kjeldahl method (AACC 1962). Protein content was obtained by multiplying the nitrogen content by 5.7.

Ash content, amylograms, farinograms, falling number, and sedimentation values were determined according to AACC approved methods (AACC 1962).

Starch damage was determined according to the method of Williams and Fegol (1969).

Free sugars were removed from flour by the method of Donovan et al (1977). Reducing sugars were assayed by the Nelson (1944) method as modified by Robyt and Whelan (1968).

Alpha-amylase activity was determined by the method of Barnes and Blakeney (1974), using the Phadebas chromogenic substrate (Pharmacia Fine Chemicals).

Exoproteolytic activity was determined by the Bushuk et al (1971) modification of the Ayre-Anderson (1939) method. Soluble nitrogen products were assayed by the ninhydrin reaction (Mertz et al 1974). Endoproteolytic activity was determined using a modified azocasein assay of Kruger (1971). Flour samples (250 mg) were suspended in 2.0 ml of 0.2*M* sodium acetate buffer (pH 6.0) and incubated with 2.0 ml of 2.5% azocasein in buffer for 2 h at 37° C in a shaking water bath. The remainder of the procedure was as described by Kruger (1971). One unit of endoproteolytic activity was defined as the change in absorbance of 0.01 after 2 h.

Free amino acid content was determined by determining the amino acids soluble in 5% sulfosalicylic acid on a Beckman model 121 automatic amino acid analyzer equipped with an Infotronic integrator, following the procedure of Spackman et al (1958).

Baking quality of flour samples was evaluated by the Grain Research Laboratory remix baking test (Irvine and McMullan 1960, Kilborn and Tipples 1981). Malt was omitted from the baking formula.

Statistical Analysis

All analyses were performed in duplicate. Analysis of variance was conducted. Least significant differences were computed at the 5% level of significance.

RESULTS AND DISCUSSION

Milling and Flour Composition

Flour yield was not greatly affected by germination (Table I). The highest flour yields were derived from the soaked wheat samples of both cultivars.

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Germination significantly altered the amounts of break and reduction flour (Table I). The yield of break flour increased to a maximum while that of reduction flour decreased with soaking and germination for both cultivars. In the milling of hard spring wheat, the proportion of break flour is normally very low; reduction flours account for most of the total flour (Ziegler and Greer 1978). Flours derived from the sound samples consisted of 20–25% break flour and 75–80% reduction flour. For both cultivars, the 18-h sample produced considerably more break flour and less reduction flour than the sound and soaked samples. Increase of germination time had no further effect on the yields of break and reduction flours. Breakdown of the kernel components make the endosperm friable such that the action of the break rolls is sufficient to reduce a greater percentage of the endosperm into flour. The yields of break and reduction flours from the soaked samples were intermediate between those from sound and 18-h samples. The imbibition of water during the soaking treatment may have modified the physical kernel structure analogous to the early changes that occur during germination. Further analytical and microscopic studies are needed to fully define the changes that occurred in kernel structure.

The composition of the flours is given in Table I. Protein content decreased slightly in both sets of samples as germination time increased, in agreement with Coulson and Sim (1965), Hwang and Bushuk (1973), and McCalla (1934). Translocation of amino acids to the developing embryo is responsible for this decrease. Ash content decreased substantially during germination and may be due to leaching and metabolic utilization of inorganic salts and mineral elements by the embryo.

Starch Degradation

Sound and soaked flour samples of both cultivars contained low α-amylase activity (Table II). Alpha-amylase activity increased significantly during germination; after 54 h, activity increased 1,600-fold and 3,000-fold for Glenlea and Neepawa, respectively.

Falling number values decreased during germination for both cultivars (Table II). Values of 60 sec for the 35- and 54-h flours indicate that the high α-amylase activity at these two treatments could not be differentiated by this method. Amylograph peak heights were zero for the 35- and 54-h samples. In both tests, values increased during the soaking stage. As α-amylase activity did not decrease during soaking, other factors, such as modification of the starch, must be responsible for this observation.

Soaking decreased the percentage of damaged starch in the milled flours, in agreement with Dronzek et al (1972) and Lineback and Ponpipom (1977) (Table II). This result may explain, in part, the observed increase in falling number values and amylograph

TABLE I
Milling Properties and Composition of Flours

Sample	Flour Yield (%)	Break Flour Yield (%)	Reduction Flour Yield (%)	Percent Protein (14% moisture)	Percent Ash (14% moisture)
Glenlea					
Sound	73.7	20	80	12.2	0.52
Soaked	75.6	51	49	11.9	0.34
18 h	72.6	67	33	12.0	0.32
35 h	72.2	67	33	11.2	0.28
54 h	73.7	68	32	11.5	0.28
LSD	Not significant	2	6	0.1	0.03
Neepawa					
Sound	74.5	25	75	11.5	0.37
Soaked	77.2	48	52	11.2	0.34
18 h	74.3	65	35	11.3	0.29
35 h	75.5	62	38	11.2	0.28
54 h	75.2	62	38	10.7	0.27
LSD	Not significant	5	7	0.3	0.04

TABLE II
Amylolytic Activity and Carbohydrate Content of Flours

Sample	α-Amylase Activity (mEU g^{-1})[a]	Falling Number Values (sec)	Amylograph Peak Height (BU)[b]	Starch Damage (%)	Reducing Sugars (mg glucose g^{-1})
Glenlea					
Sound	7	286	320	26.7	4.9
Soaked	7	335	610	12.1	4.8
18 h	477	174	70	17.5	4.9
35 h	2,944	60	0	18.1	6.0
54 h	12,000	60	0	23.9	7.4
LSD	157	22	65	5.1	1.7
Neepawa					
Sound	6	306	530	14.6	5.3
Soaked	6	365	690	11.6	5.1
18 h	348	146	10	15.2	5.1
35 h	1,296	60	10	16.7	6.2
54 h	17,500	60	0	34.0	9.5
LSD	149	8	30	2.9	0.7

[a] MEU = milli-enzyme units g^{-1}).
[b] Brabender units.

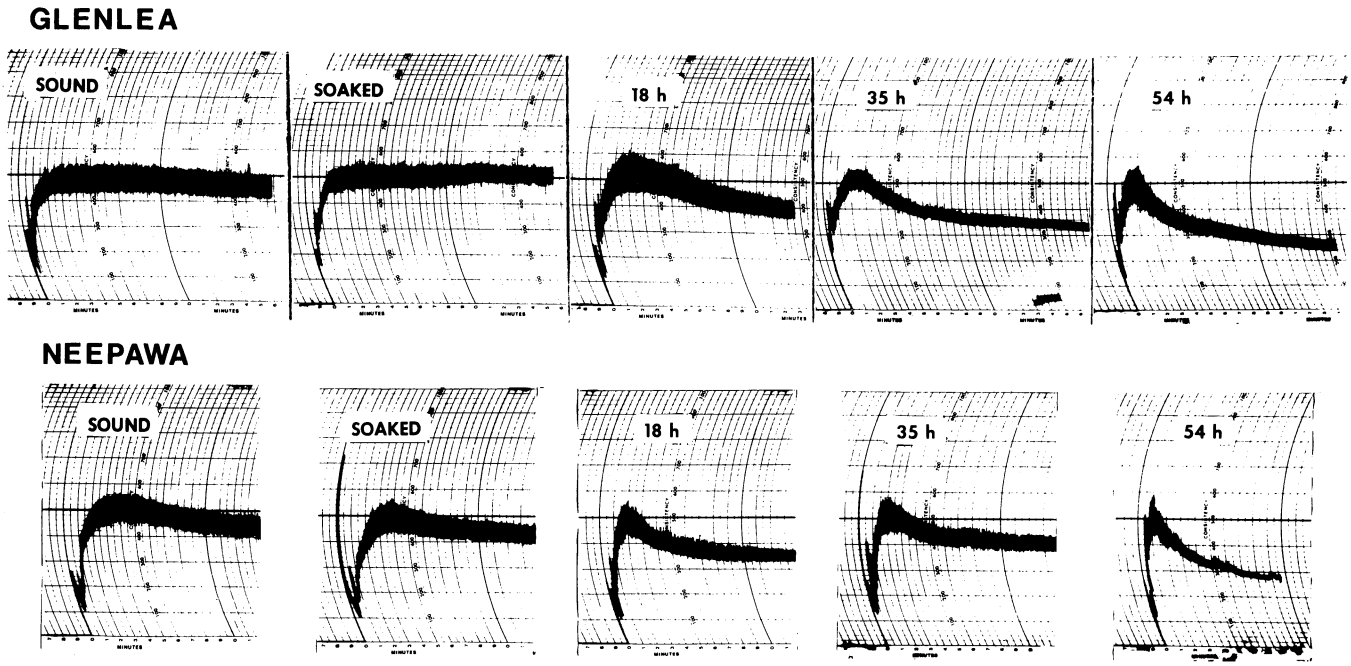

Fig. 1. Farinograms of Glenlea and Neepawa flours.

peak heights. Under a fixed set of milling conditions, the level of starch damage is directly related to kernel hardness. Results obtained for the soaked samples are consistent with the noted increase in the proportion of break flour and decrease in the proportion of reduction flour as compared to the sound flours. Generally, break flours have the lowest starch damage, and reduction flours have the highest starch damage (Holas and Tipples 1978, Pratt 1978).

With subsequent germination, the level of damaged starch increased for both cultivars, indicating that the starch was gradually degraded as germination progressed. The increase of reducing sugars during germination of both cultivars (Table II) is a further indication of enzymic degradation of starch. Furthermore, starch granules that have been partially eroded by enzyme attack are probably more susceptible to physical damage during milling.

Protein Degradation

The slight decrease of exoproteolytic activity during soaking is attributed to leaching of the enzyme (Table III). Germination produced a gradual increase in exoproteolytic activity in both cultivars; the Neepawa sample showed the largest increase after 54 h of germination (twofold). Endoproteolytic activity increased immediately at the onset of germination. The activity of the Glenlea samples increased at a faster rate than that of the Neepawa samples. After 54 h, the endoproteolytic activity of the two cultivars was essentially the same.

The increase in free amino acids during germination is a consequence of protein degradation and as such is a better indicator of total proteolytic activity than the values obtained from assays of enzyme activity on foreign substrates. Based on the values of nitrogen recovery, the total free amino acids increased approximately fourfold after 54 h of germination for both cultivars (Table III).

TABLE III
Proteolytic Activity of Flours

Sample	Exoproteolytic Activity (nmol glutamate mg^{-1})	Endoproteolytic Activity (Δ abs 0.01 $2\ h^{-1}$)	Free Amino Acids (% N recovery)
Glenlea			
Sound	61.5	3.9	0.42
Soaked	43.0	3.2	0.41
18 h	58.0	5.4	0.73
35 h	72.5	6.3	1.16
54 h	84.0	6.7	1.51
LSD	6.7	1.0	...
Neepawa			
Sound	57.0	3.6	0.50
Soaked	48.5	3.7	0.55
18 h	53.5	4.2	0.65
35 h	74.0	5.0	1.03
54 h	110.0	6.6	2.22
LSD	6.4	0.7	...

TABLE IV
Breadmaking Quality Characteristics

Sample	Sedimentation Value (cc)	Farinograph Absorption (%)	Farinograph Development Time (min)	Farinograph MTI (BU)[a]	Loaf Volume (cc)
Glenlea					
Sound	72.5	62.9	4.0	0	600
Soaked	67.0	54.2	4.0	0	630
18 h	60.5	53.5	3.0	90	813
35 h	57.0	53.1	2.5	140	603
54 h	42.0	52.6	2.0	200	433
LSD	2.0	1.3	0.2	10	66
Neepawa					
Sound	50.4	66.3	3.8	50	728
Soaked	47.4	60.3	3.8	70	438
18 h	31.0	58.0	2.0	120	615
35 h	24.9	57.7	2.0	160	460
54 h	11.0	55.4	1.0	260	395
LSD	2.6	1.3	0.1	22	69

[a] MTI = mixing-tolerance index. BU = Brabender unit.

Breadmaking Quality

Sedimentation test values for flours milled from sound wheat are directly related to breadmaking quality as assessed by loaf volume (Orth et al 1972, Pinckney et al 1957). For both cultivars, sedimentation values decreased progressively with increasing time of germination (Table IV). The values for Glenlea were consistently higher than the analogous values for Neepawa. After 54 h of germination, the Neepawa sedimentation value was less than 20, indicating exceptionally weak gluten. The corresponding Glenlea sample had a sedimentation value of 42, indicating that the gluten in this sample was still of relatively good quality.

Deterioration in baking quality was shown by the decrease in farinograph water absorption and dough development time, and increase in mixing-tolerance index during germination for both cultivars (Fig. 1 and Table IV). These results agree with those of Hwang and Bushuk (1973) and Ibrahim and D'Appolonia (1979). Quality parameters given in Table IV indicate that Glenlea can tolerate the detrimental effects of germination better than Neepawa. The farinogram of the Glenlea 18-h sample is very similar in appearance to that of the Neepawa sound sample. Dough development time decreased more rapidly for Neepawa than for Glenlea. Dough breakdown after peak development as measured by the mixing-tolerance index increased more rapidly for Neepawa than for Glenlea.

The sharp decrease in water absorption as a result of soaking is likely due to the low levels of starch damage in these samples. Although Hwang and Bushuk (1973) reported that both starch damage and farinograph absorptions decreased after soaking Manitou (a cultivar similar to Neepawa) wheat, they did not comment on a relationship between the two characteristics. Differences in water absorption between flours from sound wheat have been accounted for largely by variations in the protein and

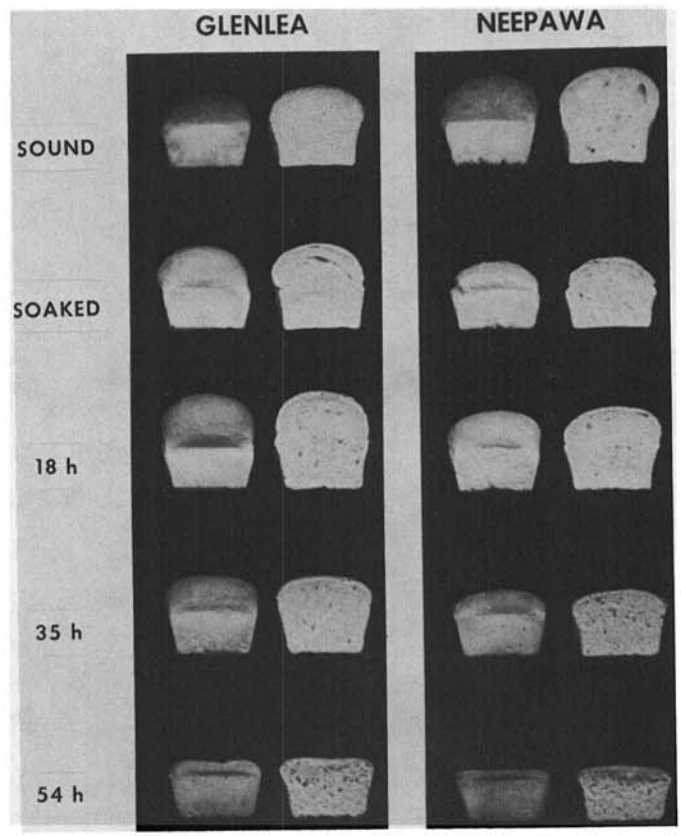

Fig. 2. External and internal loaf characteristics of the Glenlea and Neepawa samples.

damaged starch contents of the flours (Greer and Stewart 1959). The gradual decrease in absorption with germination is due to the loss of water-binding capacity of the gluten proteins, which offsets the expected rise in absorption due to the increase in the level of damaged starch.

External and internal loaf characteristics are shown in Fig. 2. Loaf volumes are given in Table IV.

For Glenlea, the 18-h flour sample produced the highest loaf volume. Dough consistency during mixing and dough-handling properties were judged to be very good. The characteristics of the loaf crust and crumb were excellent. The thin crust had a light brown color, and the crumb was white with a fine, uniform grain and satisfactory texture. Good baking properties for the Glenlea 18-h sample were anticipated from the sedimentation value and the farinograph data. As expected, the doughs of the sound and soaked Glenlea samples were bucky, indicating that they were not fully developed by the mixing process. Consequently, the baked loaves had a low volume and coarse grain. Germinating Glenlea for longer than 18 h had a detrimental effect on baking quality. Loaf volumes decreased progressively with germination. Doughs became sticky and difficult to handle, especially the 54-h sample. Crust and crumb color became darker, and crumb grain became coarser.

All treatments of Neepawa, including soaking, were detrimental to breadmaking quality, as displayed by inferior loaf volumes and crust and crumb characteristics. The Neepawa soaked sample had especially poor loaf characteristics. Soaking affected this cultivar much more than Glenlea in baking quality. The physical dough test (farinograph) and enzyme levels (amylolytic and proteolytic) do not give a clear-cut explanation of this result. Possibly, low-molecular-weight substances that were leached from the kernel are more important to baking quality for Neepawa than for Glenlea. Neepawa flour samples from germinated wheat produced progressively stickier doughs with little resistance to mixing breakdown, resulting in coarse, heavy loaves of bread.

CONCLUSIONS

Glenlea and Neepawa were similar in their responses to germination in terms of milling quality, increase in hydrolytic enzyme activity, and starch degradation. However, the changes that produced an improvement of Glenlea performance by the remix baking test produced a deterioration in the slightly weaker cultivar, Neepawa. Glenlea, with its extremely strong mixing properties, can tolerate a greater degree of germination damage than the typical red spring bread wheat cultivar, Neepawa.

Glenlea wheat has been excluded from the Canada Western red spring class because of its undesirable overly strong mixing character. The improvement of baking properties after a short period of germination presents new possibilities in the potential use of such cultivars in breadmaking.

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