Study of a Small-Scale Laboratory Wet-Milling Procedure for Wheat[1]

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ABSTRACT

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The objectives of this research were to study the effects of slurry specific gravity, starch table slope, slurry pumping rate, and their interactions on starch recovery and purity; and to propose a small-scale laboratory wet-milling procedure for wheat. First-order and second-order response surface regression models were developed to study the effects and interactions of slurry specific gravity, starch table slope, and slurry pumping rate on starch and gluten separation for a 100-g wheat wet-milling procedure. The starch and starch protein content data fit the first-order models (R^2 = 0.99 and 0.96) better than the second-order models (R^2 = 0.98 and 0.93). Regression results from the first-order models indicated that specific gravity, table slope, pumping rate, and their interactions all had a significant effect on starch yield and purity. However, these effects could be simplified as the effect of the resident time of starch and gluten slurry on the starch table and the specific gravity. Starch yield increased as resident time increased and specific gravity decreased. Protein content in starch decreased as the resident time decreased and the specific gravity increased. The separation condition with specific gravity of 3 Bé, table slope of 1.04 cm/m, and pumping rate of 50 mL/min was recommended. Under this condition, starch recovery was 85.6% and protein content of starch was 0.42%, which was similar to the 1.5-kg laboratory methods in starch recovery. Total solids recovery was 98.1%, which is similar to that from 1.5-kg laboratory methods. These results indicated that precision of the 100-g wheat wet-milling procedure was similar to that of the 1.5-kg laboratory methods.

Wheat is one of the most important crops for both human food and industry uses in the world. Currently, wheat in the United States is primarily dry-milled into flour, germ, and bran. Wheat flour consists of protein, starch, fats, fiber, and mineral matter (ash). Wheat flour from dry milling is limited in its use for end products and for its conversion to modified products. By contrast, wet-milling wheat produces starch and gluten, which can be converted into a variety of products. Starch is a basic material for food and industrial products. In food applications, starch can be chemically modified to change food texture, increase clarity, reduce gelatinization temperature, improve emulsification properties, and increase solubility. For industry uses, starch has been used in adhesives and paper coatings and as raw material for chemical fermentation into ethanol, lactic acid, polyacid and related products. Wheat gluten has been used as an additive in bread and roll formulations to improve their quality; as a fortifier of breakfast cereals; and as an extender in meat and fish products.

Separation of starch and gluten from wheat flour and wet-milling of whole wheat kernels are the two common methods to produce wheat starch and gluten. The common procedure to separate starch and gluten from wheat flour is that the flour is mixed with water to form dough and the dough is washed with water to get a starch and gluten. Weegels et al (1988) developed laboratory procedure for separation of wheat flour into starch and gluten with a 5-kg sample size. Meuser and Althoff (1989) reported that wet-milling of whole wheat kernels produced 3.5% more endosperm than dry-milling of wheat for extraction of starch and gluten. This indicates that wet-milling may have high starch and gluten recovery. Compared with processes using wheat flour, the use of whole wheat kernel also reduces damage of the starch granules (Kempf and Röhrmann 1989). Anderson (1963) developed a laboratory-scale wheat wet-milling procedure using a tabling method for starch and gluten separation. Recently, Yuan et al (1998) studied the wet-milling characteristics of hard red winter wheat using a centrifuge method for starch and gluten separation. Wet-milling quality of wheat can be evaluated by applying the above procedures, but there are some drawbacks to these methods. The wet-milling procedures described by Anderson (1963) and Yuan et al (1998) both require 1,500-g samples, and require a longer processing time. This makes them impractical for testing large numbers of wheat samples. Because of larger geographical and climatological diversity, the United States produces a huge number of wheat cultivar, and a large number of new wheat cultivars are developed each year. Therefore, there is a need to develop a small-scale laboratory testing procedure that is short and uses small quantities of grains.

To reduce processing time and labor, Eckhoff et al (1996) developed a 100-g laboratory corn wet-milling procedure that is faster and has precision similar to a 1,000-g laboratory corn wet-milling procedure. With the 100-g corn wet-milling procedure, total solids recovery and starch yields were statistically equivalent to those of 1,000-g procedure of Eckhoff et al (1993). For the 100-g corn wet-milling procedure, a 5.08-cm × 2.44-m aluminum channel was used as a starch table for separation of starch and gluten (table slope was 1.04 cm/m, specific gravity of starch and gluten slurry was adjusted into 6 Bé, and the slurry pumping rate was 50–52 mL/min). These processing parameters were optimum for corn wet-milling. However, they may not be optimum for wheat wet-milling.

Wheat has different physical and chemical properties than corn. For example, the size range of wheat starch granules (2–55 μm) is larger than that of corn starch granules (2–30 μm) (Whistler and BeMiller 1999). The large range of wheat starch granules makes separation of starch and gluten more difficult for wheat than for corn. Therefore, it is important to develop appropriate processing parameters for an improved wheat wet-milling laboratory procedure. The objectives of this research were to study the effects of slurry specific gravity, starch table slope, slurry pumping rate, and their interactions on starch recovery and purity; and to propose a small-scale laboratory wet-milling procedure for wheat.

MATERIAL AND METHODS

U.S. hard red winter wheat from the 1999 crop year was used for this research. The proximate chemical composition (%, db) of the wheat was determined by Approved Methods (AACC 2000): moisture (12.49), starch (65.3), protein (12.99), crude fiber (2.60) and ash (1.79). The test samples were first run through a Carter Day dockage tester (CEA Carter Day Co., Minneapolis, MN) to remove the foreign materials and broken kernels before steeping.

Experimental Design and Statistical Analysis

The central composite design approach (Myers and Montgomery 1995) was used to study the effects and interactions of specific gravity, table slope, and pumping rate on starch yield and purity. The central composite design is a type of response surface methodology (RSM). RSM is a general linear model in which attention is focused on characteristics of the fit response function; in particular, where optimum estimate response values occur. There were five

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levels of specific gravity (1–5 Bé), table slope (0.78–1.56 in increments of 0.26 cm/m), and pumping rate (40–60 in increments of 5 mL/min) used in these experiments (Table I). The first eight experiments (1–8) used a 2 × 2 × 2 factorial design typical for fitting the first-order models, with specific gravity of 2 and 4 Bé; table slope of 0.78 and 1.30 cm/m; and pumping rate of 45 and 55 mL/min. First-order models were then used to identify effects of specific gravity, table slope, and pumping rate on separation of starch and gluten. The last four experiments (15–18) are the center points of the design, and the six remaining experiments (9–14) are the axial points of the cube. Data from the six axial points were combined with previous data to fit the second-order models. Second-order models were also applied to identify the effects and interactions of specific gravity, table slope, and pumping rate on separation of starch and gluten. The yield data were analyzed for model fit using the response surface regression (RSREG) function (SAS Institute, Cary, NC).

Wet-Milling Procedures

The 100-g laboratory corn wet-milling procedure developed by Eckhoff et al (1996) was modified and used for this study.

Steeping. Wheat samples (100 g) were steeped in 500-mL Erlenmeyer flasks with 180-mL of steep solution with steeping temperature maintained at 38°C for 24 hr using a microprocessor-controlled water bath (model 284, Precision Co., Winchester, VA). Previous research showed that sulfur dioxide swells and disperses the protein matrix, and the degree of protein globularity is directly related to starch recovery (Cox et al 1944; Eckhoff 1983; Earp et al 1985; Yuan et al 1998). Tso (1989) showed that with additional lactic acid in steepwater, wet-milling yield and product quality were improved. Yuan et al (1998) used the lactic acid as steeping agent for wheat wet-milling. The steeping solution with 0.3% (w/w) sulfur dioxide and 0.5% (w/w) lactic acid was used in this study. After steeping, the unabsorbed steepwater was drained into a 250-mL graduated cylinder and measured. Steepwater was then transferred into a 50-mL aluminum cap and dried in a forced-air oven at 49°C for 24 hr to remove most of the water. It was then transferred to another oven and dried at 103°C for 5 hr (AACC 2000).

First grind. The steeped wheat was ground in 200 mL of distilled water at 15,000 rpm for 1 min using a Waring-type blender (model CB-6, Dynamic Corp. of America, New Hartford, CT). The blender was equipped with a removable 1-L stainless steel container and stainless steel blades.

Second grind. After first grind, the stainless steel container with the coarse wheat slurry was removed from the blender. The coarse slurry was then second ground using a plate mill (Quaker City model 4E, Straub Co., Hatboro, PA). The desired fineness of grind was achieved by adjusting plate pressure. Water (≈200 mL) was used to wash the blender and plate mill.

Fiber separation. Fiber and germ were separated from the starch and gluten slurry using a standard testing sieve (U.S. No. 100, 150 µm). The sieve was mounted on the top of a 7.5-L bucket and then this sieve and bucket arrangement was mounted on a Ro-Tap shaker (model RX-29, W.S. Tyler Inc., Montor, OH). During shaking (≈5 min), the slurry was slowly poured on the sieve and ≈500 mL of distilled water was used to wash the materials on the sieve. The mixture of fiber and germ retained on the sieve was transferred onto a weighted tray and dried using the two-step air-oven procedure to determine the recovery of fiber and germ (AACC 2000).

TABLE I
Central Composite Design for Three Variables at Five Levels

	Coded Variables			Actual Experimental Variables[a]		
Experiment	X_1	X_2	X_3	SG	TS	PR
1	–1	–1	–1	2.0	0.78	45
2	+1	–1	–1	4.0	0.78	45
3	–1	+1	–1	2.0	1.30	45
4	+1	+1	–1	4.0	1.30	45
5	–1	–1	+1	2.0	0.78	55
6	+1	–1	+1	4.0	0.78	55
7	–1	+1	+1	2.0	1.30	55
8	+1	+1	+1	4.0	1.30	55
9	–2	0	0	1.0	1.04	50
10	+2	0	0	5.0	1.04	50
11	0	–2	0	3.0	0.52	50
12	0	+2	0	3.0	1.56	50
13	0	0	–2	3.0	1.04	40
14	0	0	+2	3.0	1.04	60
15	0	0	0	3.0	1.04	50
16	0	0	0	3.0	1.04	50
17	0	0	0	3.0	1.04	50
18	0	0	0	3.0	1.04	50

[a] SG = specific gravity of starch and gluten slurry (Bé); TS = table slope (cm/m); PR = pumping rate (mL/min).

TABLE II
Parameters[a] of the First-Order Regression Models for Starch Yield and Protein Content in Starch[b]

Parameters	Starch (%)	Protein (%)
β_0 (intercept)	91.157	3.174
β_1 (TS)	–20.765	–1.775
β_2 (PR)	–0.551	–0.045
β_3 (SG)	–3.900	–0.175
β_4 (TS × PR)	0.209	0.026
β_5 (SG × PR)	0.044	0.003
β_6 (SG × TS)	–0.445	0.020

[a] SG = specific gravity of starch and gluten slurry, TS = table slope (cm/m), PR = pumping rate (mL/min).

[b] Starch yield/protein content in starch = $\beta_0 + \beta_1 \times \text{TS} + \beta_2 \times \text{PR} + \beta_3 \times \text{SG} + \beta_4 \times \text{TS} \times \text{PR} + \beta_5 \times \text{SG} \times \text{PR} + \beta_6 \times \text{SG} \times \text{TS}$.

TABLE III
Significance of the First-Order Regression Model Parameters for Starch Yield and Protein Content (%) in Starch

	Starch Yield			Protein Content in Starch		
Regression	*F* Value	*Pr* > *F* or *T*	R^2	*F* Value	*Pr* > *F* or *T*	R^2
Model						
Linear	483.9	0.0001	0.97	193.8	0.0001	0.90
Crossproduct	8.0	0.0066	0.02	17.8	0.0004	0.08
Total	246.0	0.0001	0.99	105.8	0.0001	0.98
Lack of fit	8.0	0.0223	···	···	11.6	0.009
Variables[a]						
Intercept	···	0.0001	···	···	0.0001	···
TS	···	0.0001	···	···	0.0001	···
PR	···	0.0001	···	···	0.0001	···
SG	···	0.0008	···	···	0.0090	···
TS × PR	···	0.0060	···	···	0.0001	···
SG × PR	···	0.0151	···	···	0.0201	···
SG × TS	···	0.1625	···	···	0.3320	···

[a] SG = specific gravity of starch and gluten slurry; TS = table slope (cm/m); PR = pumping rate (mL/min).

Starch-gluten separation. The separation of starch and gluten was achieved using 5.08-cm × 2.44-m aluminum starch tables. The empty starch table was weighed before tabling. The combinations of the table slope, specific gravity, and pumping rate for starch and gluten separation were listed in Table I and were detailed earlier. The desirable specific gravity was adjusted by decanting from the starch and gluten mixture or adding distilled water into the mixture. The mixture of the starch and gluten was pumped onto starch table by using MasterFlex pump (508997, Cole-Parmer Instrument Co., Chicago, IL). After tabling the starch and gluten slurry and the decanted water, ≈150 mL of distilled water was used to wash the bucket and remove gluten residual from the starch table. Starch was then ambient-dried overnight on the starch table. On the second day, the starch table was weighed and starch was collected. Starch (≈35 g) was used for moisture content determination using a forced-air oven at 135°C for 2-hr.

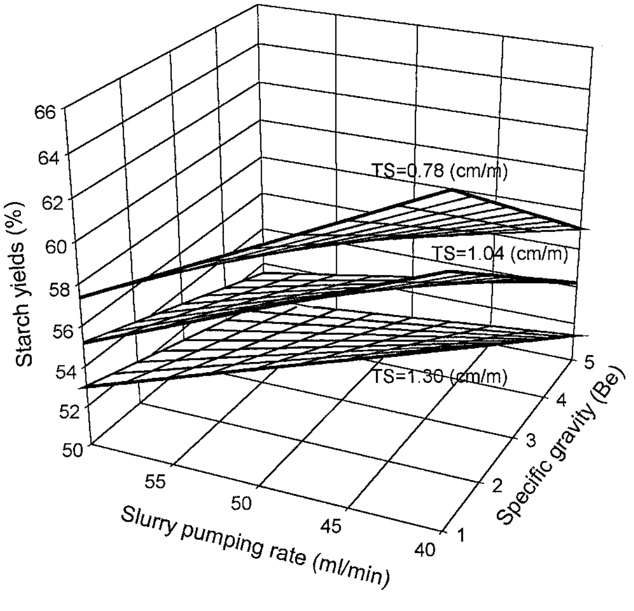

Fig. 1. Effects of slurry pumping rate and specific gravity on starch yields at a constant table slope (TS).

Gluten centrifugation. The overflow from starch tabling was transferred to 800-mL centrifuge bottles and centrifuged for 10 min at 4,000 rpm (model J2-21 Beckman). After centrifugation, the processing water was decanted, and the gluten was scraped. The gluten was dried using the two-stage air-oven method for determination of gluten recovery (AACC 2000). Three 30-mL samples of processing water were used to determine the solid content in the processing water using the same procedure described earlier for determination of steepwater solid content (AACC 2000).

RESULTS AND DISCUSSION

Regression Models

The first-order and second-order response surface models were developed to study the effects of slurry specific gravity, starch table slope, slurry pumping rate, and their interactions on starch recovery and purity. Because the starch yield and starch protein content data fit the first-order models ($R^2 = 0.99$ and 0.96) better than the second-order models ($R^2 = 0.98$ and 0.93). The second-order regression results were not reported.

The first-order regression models describe the linear influences of specific gravity, table slope, pumping rate, and their interactions on starch yield and purity. General first-order regression model is:

$$\hat{y} = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_1X_2 + \beta_5 X_1X_3 + \beta_6 X_2X_3 \quad (1)$$

where $\hat{y}$ is starch yield or protein content in starch, X_1 is table slope (cm/m), X_2 is pumping rate (mL/min), X_3 is specific gravity (Bé), and $\beta_0 \ldots \beta_6$ are intercept and estimated coefficients (Table II). Significance of regression models and individual variables were determined by calculating the 95% confidence intervals. The first-order regression models for starch yield and protein content in starch were significant ($P < 0.05$) (Table III). The lack-of-fit was also significant at $P < 0.05$ levels.

Based on results from the first-order model, all three variables significantly affected starch yield ($P < 0.05$) for both linear and crossproduct model fit. Coefficient of determination showed that all three variables were significant predictors of starch yield ($R^2 = 0.99$) (Table III).

The table slope influences the resident time of the slurry on the starch table and there was a negative relationship between table slope and resident time. With a small table slope, resident time of

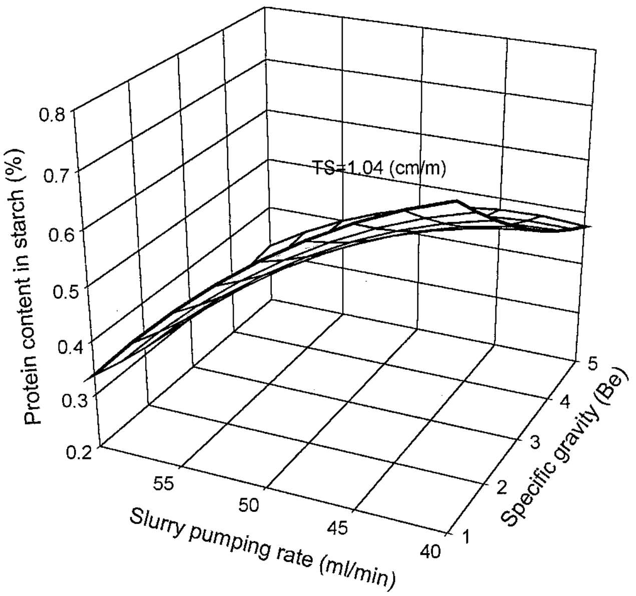

Fig. 2. Effects of slurry pumping rate and specific gravity on starch protein content at a constant table slope (TS).

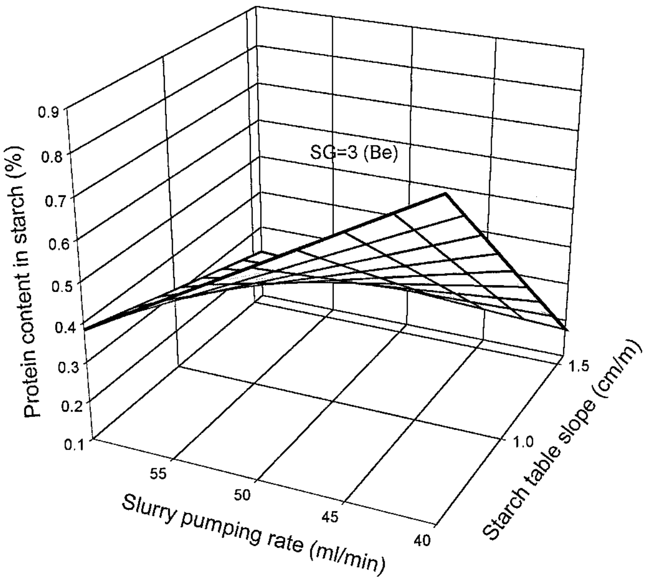

Fig. 3. Effects of slurry pumping rate and starch table slope on starch protein content at a constant specific gravity (SG).

slurry on the table was longer than with a larger table slope. With a longer resident time, the starch granules had enough time to fall down. As a result, starch yield increased as table slope decreased at a constant pumping rate and specific gravity (Fig. 1). Figure 1 also shows the effect of pumping rate and specific gravity on starch yield. At a constant specific gravity and table slope, the starch yield decreased as pumping rate increased. This is because pumping rate also affects resident time of slurry on the starch table. With a high pumping rate, the starch granules did not have enough time to fall down on the starch table, especially for the smaller size starch granules. At a constant pumping rate and table slope, the starch yield decreased as specific gravity increased. With a high specific gravity, some larger starch granules were floated with gluten in the overflow, resulting in a low starch yield. The highest starch yield occurred at the lowest values for both pumping rate and specific gravity and the lowest starch yield occurred at the highest values for both pumping rate and specific gravity. Similar patterns were found for the effect of specific gravity and table slope with a constant pumping rate and the effect of table slope and pumping rate with a constant specific gravity. These effects could be simplified as the effects of the resident time of slurry on the starch table and specific gravity. The starch yield increased as resident time increased and specific gravity decreased. However, because of the limitation of protein content in starch, separation conditions that resulted in the highest starch yield may be not the optimum processing conditions.

Specific gravity, table slope, and pumping rate were significant factors affecting protein content of starch, based on the first-order model for protein content ($R^2 = 0.98$, $P < 0.05$ for both linear and crossproduct model fit) (Table III). In addition, significant interactions were noted for table slope and pumping rate and for specific gravity and pumping rate ($P < 0.05$).

In general, specific gravity, table slope, and pumping rate had similar effects and patterns on protein content of starch as they did for starch yield. Figure 2 shows the effect of the pumping rate and specific gravity on protein content of starch at a constant table slope. Protein content increased as pumping rate and specific gravity decreased. This is because, at a constant specific gravity, decreasing pumping rate increased the resident time of slurry on the starch table and some small gluten particles fell down on the starch table resulting in higher protein content. The protein content increased as the specific gravity decreased. When specific gravity was small, smaller gluten particles fell down easily onto the starch table, resulting in higher protein content in starch. However, at the high pumping rate, specific gravity had no effect on starch protein content (Fig. 2). Pumping rate also had no significant effect on protein content at the larger table slope (1.56 cm/m) (Fig. 3). After the resident time was reduced beyond a certain level, there was no additional effect on protein content in starch, only on starch yield.

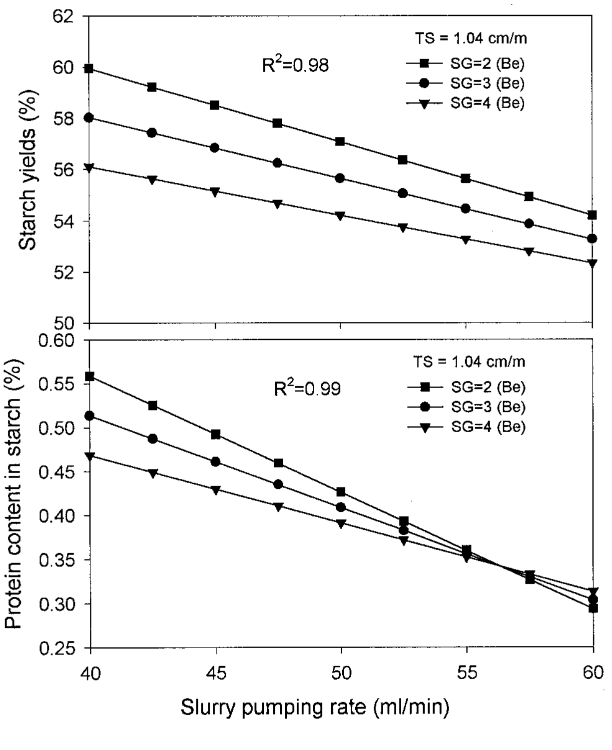

Fig. 4. Effect of slurry pumping rate and specific gravity (SG) on starch yields and starch protein content at a constant table slope (TS).

Selecting an appropriate processing condition for starch and gluten separation should meet the criteria of a low level of the protein content in starch as well as an adequate starch yield. For industrial applications, protein content in starch should be <0.5%. Figure 4 shows the relationship between starch yield and protein content in starch when the table slope was kept constant (1.04 cm/m). Protein content in starch was >0.4% when the pumping rate was <50 mL/min for specific gravity levels of 2 and 3 Bé. When the pumping rate was increased from 50 to 57.5 mL/min, starch yield was reduced by 2.5–5.0%. This result indicated that the optimum pumping rate should be ≈50 mL/min, and a specific gravity of 3 Bé is recommended for a table slope of 1.04 cm/m.

Comparison with Previous Study

The wet-milling yields from the 100-g procedure were compared with previous results (Anderson 1963; Kempf and Röhrmann 1989; Yuan et al 1998) (Table IV). A 100-g procedure with limited slurry transfer from container to container minimized processing losses in our study. The total solid recovery of 98.1% was similar to the results from the 1.5-kg wet-milling procedure described by Anderson (1963). Results from our 100-g procedure (85.6%) were also close to the starch recovery reported by Yuan et al (1998) (86.1%). Results were >10% of those reported by Anderson (1963) (76%), and >6% than those reported by Kempf and Röhrmann (1989) (80%). The 100-g procedure yielded starch with protein content of 0.42%, which was the same as the results obtained by Kempf and Röhrmann (1989). However, this result is higher than that reported by either Yuan et al (1998) or Anderson (1963) (0.34 and 0.22%, respectively). Even though protein content was greater in our 100-g procedure, which could be a disadvantage of this shorter, small-scale procedure, protein content <0.5% was considered acceptable.

TABLE IV
Comparisons of Wet-Milling Yields Among Four Studies

	100-g Procedure	Yuan et al (1998)	Anderson (1963)	Kempf and Röhrmann (1989)
Raw material[a]	HRW	HRW	SWW	HRS
Capacity, kg/batch	0.1	1.5	1.5	···
Steeping condition				
SO_2 (%)	0.3	0.3	0.3	0.3
Lactic acid (%)	0.55	···	···	···
Temperature (°C)	38.0	37.0	38.0	38.0
Steeping time (hr)	24.0	24.0	24.0	24.0
Yields				
Starch				
Yield (%)	55.8	56.1	51.2	54.0
Recovery (%)	85.6	86.1	76.0	80.8
Protein content (%)	0.42	0.34	0.22	0.42
Gluten (%)	21.5	16.1	22.4	22.3
Fiber (%)[b]	11.5	19.3	12.3	···
Steepwater solids (%)	2.8	···	2.9	···
Processing water solids (%)	6.5	···	9.4	···
Total dry matter recovery (%)	98.1	91.5	98.2	···

[a] HRW = hard red winter; SWW = soft white winter; HRS = hard red spring.
[b] Fiber includes both fiber and germ.

Gluten yield from the 100-g procedure was similar to results from Anderson (1963) and Kempf and Röhrmann (1989), but was higher than results from Yuan et al (1998). This is probably because fiber yield from the 100-g procedure was lower than fiber yield from Yuan et al (1998) and similar to the fiber yield from Anderson (1963). The steeping water solids content obtained from the 100-g procedure and 1.5-kg laboratory method (Anderson 1963) was the same. However, the processing water solids content from Anderson (1963) was much higher than that from our 100-g procedure. With larger sample, more processing water was used, thereby generating more processing water solids.

CONCLUSIONS

The starch yield and starch protein content data fit the first-order regression models with R^2 values of 0.99 and 0.96, respectively. The effect of specific gravity, table slope, and pumping rate, and their interactions on the starch yield and purity could be simplified as the effect of resident time of slurry on the starch table and specific gravity. The starch yield increased as the resident time increased and the specific gravity decreased. protein content decreased as the resident time decreased and specific gravity increased. Separation conditions with a specific gravity of 3 Bé, table slope of 1.04 cm/m, and pumping rate of 50 mL/min yielded best results. The starch recovery of 85.6% and protein content of starch of 0.42% were achieved under these conditions. Our results for starch recovery were similar to laboratory methods using 1.5-kg samples. Also, results indicated that the precision of the 100-g wheat wet-milling procedure was similar to that achieved with 1.5-kg laboratory methods.

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