Hydrocyclone Procedure for Starch-Protein Separation in Laboratory Wet Milling

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ABSTRACT

A hydrocyclone system for starch-protein separation was developed for use with 1-kg samples in laboratory corn wet milling. A Doxie 5 hydrocyclone with all but one cyclone plugged and a five-pass starch washing system was compared to a traditional starch tabling procedure using both regular dent and waxy corn hybrids. The tabling procedure gave 3–4% higher starch yields in dent corn and 2–3% higher starch yields in waxy corn. Tabled starch had less protein (0.33 and 0.45% for dent and waxy, respectively) than the Doxie 5 hydrocyclone-separated starch (0.64 and 0.65% for dent and waxy, respectively). Using a Doxie Type A single hydrocyclone instead of the Doxie 5 increased the starch yield; however, protein in starch increased to 1.29 and 0.97% for dent and waxy, respectively. Design and operational differences may account for the different results. The hydrocyclone procedure reduced the time required for starch-protein separation by 75%. It also eliminated the requirement of a large floor area for starch tables, reduced the potential for operator error, and more closely simulated the starch-protein separation process used in industrial operations. The reduced testing time and ease of use will make the hydrocyclone procedure useful for comparing milling procedures or different corn hybrids.

Laboratory and pilot-scale wet milling procedures can be used to evaluate the wet milling characteristics of new corn hybrids, the effect of harvest and postharvest processing methods on corn millability, and the use of different steeping and processing techniques on product yields. A laboratory-scale study can be conducted when a pilot-scale study would be too expensive or when only a limited amount of a particular hybrid is available for testing.

Traditional laboratory-scale corn wet milling procedures presented in the research literature (Watson et al 1951, Anderson 1963, Steinke and Johnson 1991, Eckhoff et al 1993) use either starch tabling or batch centrifugation for starch-protein separation. However, in industrial operations, corn is processed by continuous centrifuges and a system of hydrocyclone separators that repeatedly refine the starch-protein using 2.1–2.5 L of water per kilogram of dry starch (May 1987). Rubens (1990) developed a pilot-scale wet milling facility using four 10-mm hydrocyclones (TM 1 and TM 3, Dorr-Oliver, Inc., Stamford, CT). But pilot-scale wet milling requires large quantities of corn and is practical only when large quantities of starch are required for research or modification purposes.

The objectives of this study were: 1) to establish a starch-protein separation procedure for 1-kg samples using a hydrocyclone; 2) to compare the resulting starch fractions with those obtained using a traditional starch tabling procedure; 3) to compare the performance of two commercially available 10-mm hydrocyclones (Doxie 5 and Doxie Type A, Dorr-Oliver).

MATERIALS AND METHODS

Reproducibility Tests

To check the reproducibility of a laboratory starch-protein separation technique using hydrocyclones, samples of the corn were fractionated into steepwater, germ, and fiber using the laboratory wet milling procedure of Eckhoff et al (1993). Starch-protein separation was obtained by five passes through a hydrocyclone system in place of a starch table.

A blend of ambient air-dried commercial waxy hybrids available from a concurrent project was used. Ten 1-kg samples were steeped in 2,000 ml of steeping solution containing 0.3% sodium metabisulfite (1,500 ppm SO_2) and 0.55% lactic acid at 50°C for 24 hr.

The hydrocyclone system (Fig. 1) consisted of a 10-mm hydrocyclone (Doxie 5) connected to a positive displacement pump (Hydra Cell, Wanner Engineering, Minneapolis, MN) fed by an overhead tank. The Doxie 5 unit consisted of six 10-mm hydrocyclones in parallel operation. Five of these hydrocyclones were plugged using two rubber stoppers for each cyclonette to reduce feed requirement and make it suitable for laboratory-scale operation. Hydrocyclone inlet pressure was controlled by recirculating part of the pump capacity back to the overhead tank. Both the underflow and the overflow of the hydrocyclone were valved, but the valves were kept open during the testing.

After fiber separation, the mill starch slurry was adjusted with fresh water to a density of 1–2° baume (sp gr 1.0069–1.0140). This mill starch was then added to the overhead hydrocyclone tank. The hydrocyclone inlet pressure was adjusted to 8.96–9.65 bar (130–140 psi). Preliminary studies showed that pressures >9.65 bar (140 psi) would throw larger sized protein particles into the starch fraction; pressures <8.96 bar (130 psi) would throw smaller sized starch particles into the protein fraction. The starch slurry obtained at the underflow was mixed with 4,000 ml of water and passed again through the hydrocyclone to wash and clean the starch. Five washing passes were used to clean the starch.

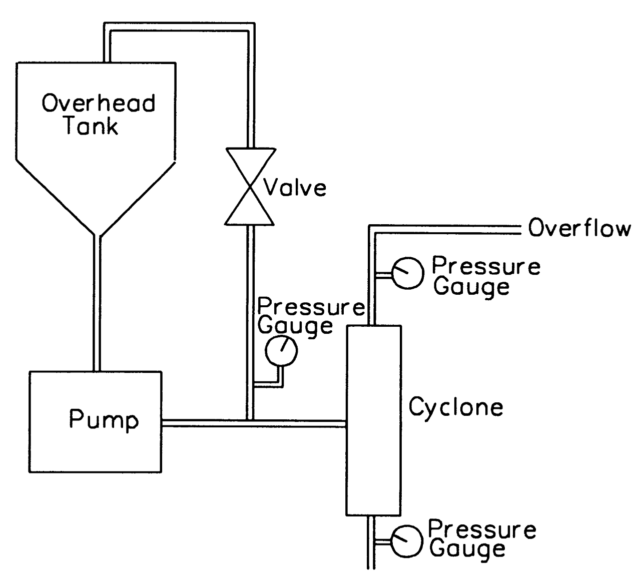

Fig. 1. Schematic diagram of hydrocyclone used for starch-protein separation.

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Preliminary studies indicated that five washings was a compromise between cleaner starch with more washings and the associated starch yield loss. Starch going to the overflow in the protein fraction at any stage was not recovered.

Underflow from the hydrocyclone, the starch fraction, was dewatered by vacuum filtering at ~560–660 mmHg through a 25-cm Buchner funnel using 24-cm (type 1) Whatman filter paper. The liquid that passed through the vacuum filter was recovered and repassed through the same Buchner funnel to enhance starch recovery. The second-pass liquid was checked for residual solids. Residual solids were negligible, and the second-pass liquid was discarded.

The volume of the protein fraction (overflow from all passes of the hydrocyclone), was measured. Five 50-ml aliquots were dried in a two-stage forced-air oven to determine solids content (AACC 1983).

Milled fractions (steepwater, germ, fiber, starch, and protein) were dried at 49°C for 24 hr in a forced-air convection-drying oven to stabilize the fractions, which were then weighed. A small sample of all solid milling fractions (germ, fiber, and starch) was dried at 103°C for 2 hr to determine the moisture content. Aliquots of the liquid milling fractions (steepwater and protein) were dried at 103°C for 5 hr to determine the solids content of these fractions (AACC 1983).

Comparison Between Tabling and Hydrocyclone Separation Methods

To make a comparative evaluation of the starch-protein separation between hydrocyclone and starch tabling methods, blends of both a regular dent corn and a commercial waxy corn hybrid were wet milled in triplicate. The blends of regular dent corn were obtained by mixing equal amounts (by weight) of three hybrids: FR27×FRMo17 (a soft endosperm corn), FR618×FR600 (a medium-hard endosperm corn), and FR1087×LH123 (a hard endosperm corn). These hybrids were obtained from the 1993 harvest of the agricultural engineering farm at the University of Illinois at Urbana-Champaign. Hybrids were ambient air-dried in the laboratory. The blends of commercial ambient air-dried waxy corn hybrids were procured from a local corn processing company.

In addition to the Doxie 5 unit, a single 10-mm hydrocyclone (Doxie Type A) was also tested. The Doxie Type A unit consisted of only one 10-mm hydrocyclone, thus eliminating the need of plugging any hydrocyclone. Three replicates each of regular dent corn blend and waxy corn blend were milled using this hydrocyclone for starch-protein separation.

Starch-protein content (% protein = % nitrogen × 6.25) was determined using a Kjeldahl nitrogen procedure by block digestion and steam distillation (CRA 1980) at an outside analytical laboratory. The particle-size distribution of the starch powder obtained by the tabling and the Doxie 5 methods were compared. Light scattering analysis (LA-900, Horiba Instruments, Irvine, CA) performed at an outside analytical laboratory was used to obtain particle-size distribution of starch powders.

Statistical software (SAS Institute, Cary, NC) was used to analyze the data. One-way analysis of variance (ANOVA) was used to determine the significant differences in the two procedures for starch-protein separation. The probability of α (type I error) was 1% ($P < 0.01$).

RESULTS AND DISCUSSION

Reproducibility of the Hydrocyclone Procedure

The waxy corn milled in this study to validate the reproducibility of the hydrocyclone procedure exhibited typical laboratory milling yields (yield refers to fraction of corn component expressed as % dsb of initial dry corn solids), with germ, fiber, starch, and protein yields of ~7, 13, 61, and 14% (dsb), respectively (Table I). Sufficient steeping is indicated by relatively low fiber and gluten yields and by low protein content in starch. High recovery rates (total corn components recovered expressed as %dsb of initial dry corn solids) and low protein contents in starch were observed for the procedure. The relatively low coefficient of variation (CV) indicated that reproducible results can be obtained using this procedure.

The relatively high CV for steepwater solids (10.0%) and protein yields (9.2%) is attributable to aliquot sampling error. Drying of the whole liquid fractions is not feasible due to the large amounts of liquids. The CV of starch yields and percentage recovery are very low (<1%) because they have high mean values.

Variability in germ and fiber yields is attributable to operator error during manual operations. Time spent for germ skimming and fiber washing was not controlled. However, the CV for germ and fiber yields (~4%) is reasonable. Protein content in starch has a CV of 6.42% even though the values are very close to each other because of the small mean value.

Comparison of Fractions Obtained from Tabling and Hydrocyclone Separation Methods

ANOVA conducted for the two procedures (tabling and Doxie 5) for regular dent corn (Table II) showed that the steepwater solids and the germ and fiber yields were not significantly different for the two procedures. The starch and protein yields, and the protein content in starch were significantly different.

The starch yields obtained from starch tabling (66.61%) were higher than the starch yields obtained from the Doxie 5 unit (62.57%). Because hydrocyclones tend to be classifying rather than clarifying devices, some starch is discharged in the overflow (pro-

TABLE I
Wet Milling Yields of Waxy Corn Components and the Protein Content in Starch for 10 Replicate Determinations of the Hydrocyclone Procedure[a]

Replicate	Steepwater	Germ	Fiber	Starch	Protein	Recovery[b]	Starch Protein Content
1	3.02	6.59	11.99	61.74	14.44	97.78	0.81
2	3.05	6.98	12.98	59.61	14.31	96.93	0.76
3	3.07	6.55	13.38	61.07	12.74	96.80	0.79
4	3.44	6.68	13.48	60.96	12.38	96.94	0.80
5	3.33	6.45	13.53	60.87	13.04	97.22	0.71
6	2.94	6.61	13.29	60.75	13.08	96.67	0.66
7	3.25	6.18	12.65	60.78	16.33	99.22	0.73
8	3.75	7.14	13.05	60.63	13.62	98.19	0.80
9	2.64	6.75	13.28	60.70	14.42	97.78	0.76
10	3.50	6.44	12.66	60.24	15.72	98.58	0.80
Average	3.20	6.64	13.03	60.73	14.01	97.61	0.76
SD[c]	0.32	0.27	0.48	0.55	1.29	0.85	0.05
CV, %[d]	10.00	4.12	3.69	0.90	9.22	0.87	6.42

[a] Fractions of corn components and protein content in starch expressed as % dry solids basis.
[b] Percentage recovery of initial dry corn solids.
[c] Standard deviation.
[d] Coefficient of variation.

tein fraction) with each pass (Bier 1983). The overflow from the five-pass washing of the hydrocyclone procedure was primarily protein and was not recirculated through the hydrocyclone at any stage. With no secondary recovery of starch in the overflow of five passes, lower starch yields were expected.

The protein content of the starch obtained using the Doxie 5 unit (0.64%) was higher than the protein content in starch from tabling (0.33%). The five passes of the hydrocyclone starch washing system reduced the protein content in starch, but simultaneously reduced starch yields by forcing more starch into the overflow.

ANOVA conducted for the tabling and the Doxie Type A data showed that the steepwater solids and the germ, fiber, starch and protein yields were not significantly different for the two procedures. However, the protein content in starch was significantly different. When the Doxie Type A unit was used in place of the Doxie 5, the starch yield and the protein content in starch were significantly increased. The Doxie 5 hydrocyclone has a strainer, whereas the Doxie Type A does not. The feed for Doxie 5 hydrocyclone is introduced into the center of the strainer and has to pass through the strainer before it enters the hydrocyclones.

The operating pressure drop characteristics for the Doxie 5 and Doxie Type A units are similar (Dorr-Oliver 1992). The diameter of the Doxie 5 feed, underflow, and overflow are each 12.7 mm (0.5 in.). These dimensions are larger than those of the Doxie Type A feed, underflow, and overflow (9.5, 9.5, and 3.2 mm [0.375, 0.375, and 0.125 in.], respectively). Decrease in feed diameter reduces the capacity of the system (Bradley 1965):

$$Q \propto D_i^z$$

where Q is capacity, D_i is inlet diameter, and z is an empirical constant with a value in the 0.9–2.0 range. Furthermore, smaller feed diameter leads to higher separation efficiency and affects the cut size appreciably (majority of the particles finer than the cut size in the feed go to overflow, while the majority of the coarser particles are separated as underflow) (Svarovsky 1984). Bradley (1965) summarized the empirical relationships governing cut size and feed diameter as:

$$d_{50} \propto D_i^x$$

where d_{50} is the diameter of the particle exhibiting a centrifugal efficiency of 50%, D_i is inlet diameter, and x is an empirical constant with a value in the 0.6–0.68 range. A reduction in feed diameter lowers the cut size. To increase the cut size, Doxie Type A hydrocyclones should be operated at a lower differential pressure. These factors implied that operating parameters (differential pressure and the number of washing passes) should be different for the Doxie Type A and the Doxie 5 hydrocyclones.

In this study, the Doxie Type A hydrocyclone was operated at the same pressure as the Doxie 5 hydrocyclone. The higher pressure forced the smaller sized starch particles and larger sized protein particles toward the walls of the cyclone and finally in the underflow (starch fraction), resulting in higher starch yield and higher protein content in starch.

Wet milling a blend of waxy corn using either a hydrocyclone or a starch table for starch-protein separation showed results that were similar to those obtained by wet milling regular dent corn (Table III). ANOVA conducted for waxy corn showed that the steepwater solids and the germ and fiber yields were not significantly different for the two procedures (tabling and Doxie 5 unit), whereas the starch and protein yields and the protein content in starch were significantly different. The starch yields obtained from tabling (61.77%) were higher than the starch yields obtained using a Doxie 5 unit (59.05%). The protein content of the starch obtained using the Doxie 5 unit (0.66%) was higher than the protein content in starch obtained from tabling (0.45%). ANOVA conducted for the tabling and the Doxie Type A procedures showed that the steepwater solids and the germ, fiber, starch, and protein yields were not significantly different. However, the protein content in starch was significantly different for the two procedures.

Starch yields obtained from the Doxie 5 unit are closer to those obtained from tabling for waxy corn than they were to those for regular dent corn. Waxy corn has a larger volume percentage of median-sized starch particles when compared to regular dent corn. These median-sized starch particles end up in the underflow. The result is that, for waxy corn, starch yields from the Doxie 5 procedure are closer to those obtained from tabling.

Although the tabling procedure gave higher starch yields and lower protein content in the starch when compared to the Doxie 5 unit, the hydrocyclone procedure showed other advantages over tabling. The time required for starch-protein separation using the hydrocyclone was ~75% lower when compared with starch tabling. Setting up both the hydrocyclone and the starch table (including peristaltic pump) at the laboratory scale requires a capital investment of $1,000–1,500. However, because the hydrocyclone procedure requires less time, operational costs are reduced

TABLE II
Wet Milling Product Yields and Starch-Protein Content for Regular Dent Corn[a]

Replicate	Steepwater	Germ	Fiber	Starch	Protein	Recovery[b]	Starch Protein Content
Tabling method							
1	3.17	6.74	10.79	66.75	11.82	99.26	0.34
2	3.38	6.99	10.91	66.80	11.21	99.29	0.33
3	2.90	6.74	11.07	66.28	12.79	99.78	0.33
Average	3.15 a[c]	6.82 a	10.92 a	66.61 a	11.94 a	99.44 a	0.33 a
SD[d]	0.19	0.12	0.12	0.24	0.65	0.24	0.01
Hydrocyclone (Doxie 5) method							
1	3.71	6.78	11.05	62.09	15.50	99.13	0.57
2	3.43	6.18	11.27	62.49	15.57	98.92	0.64
3	2.99	6.73	11.28	63.12	15.27	99.39	0.70
Average	3.38 a	6.56 a	11.20 a	62.57 b	15.44 b	99.15 a	0.64 b
SD	0.29	0.27	0.11	0.42	0.13	0.19	0.05
Hydrocyclone (Doxie Type A) method							
1	3.60	6.48	10.77	65.95	11.97	98.77	1.23
2	3.55	6.28	11.04	66.01	12.07	98.95	1.27
3	3.56	6.43	10.80	65.49	12.47	98.73	1.36
Average	3.57 a	6.40 a	10.87 a	65.81 a	12.17 a	98.81 a	1.29 c
SD	0.02	0.08	0.12	0.23	0.22	0.09	0.05

[a] Fractions of corn components and protein content in starch expressed as % dry solids basis.
[b] Percentage recovery of initial dry corn solids.
[c] Means followed by the same letter in the same column are not significantly different at $\alpha = 0.01$.
[d] Standard deviation.

TABLE III
Wet Milling Product Yields and Starch Protein Content for Waxy Corn[a]

Replicate	Steepwater	Germ	Fiber	Starch	Protein	Recovery[b]	Starch Protein Content
Tabling method							
1	3.54	6.49	11.43	61.96	14.98	98.40	0.43
2	4.91	6.03	11.60	61.66	14.62	98.83	0.45
3	4.66	6.27	11.16	61.68	15.16	98.93	0.48
Average	4.37 a[c]	6.26 a	11.40 a	61.77 a	14.92 a	98.72 a	0.45 a
SD[d]	0.59	0.19	0.18	0.14	0.22	0.23	0.02
Hydrocyclone (Doxie 5) method							
1	3.48	6.43	12.77	58.98	16.64	98.30	0.64
2	4.71	6.34	11.45	59.00	16.53	98.03	0.67,
3	4.94	6.82	11.76	59.15	16.25	98.92	0.68
Average	4.38 a	6.53 a	11.99 a	59.05 b	16.47 b	98.42 a	0.66 b
SD	0.64	0.21	0.56	0.08	0.16	0.37	0.02
Hydrocyclone (Doxie Type A) method							
1	4.77	6.26	11.31	62.13	14.27	98.74	0.95
2	4.70	6.53	11.14	61.97	14.23	98.58	0.94
3	4.75	6.35	11.32	62.05	14.11	98.58	1.01
Average	4.74 a	6.38 a	11.26 a	62.05 a	14.20 a	98.63 a	0.97 c
SD	0.03	0.11	0.08	0.07	0.07	0.08	0.03

[a] Fractions of corn components and protein content in starch expressed as % dry solids basis.
[b] Percentage recovery of initial dry corn solids.
[c] Means followed by the same letter in the same column are not significantly different at $\alpha = 0.01$.
[d] Standard deviation.

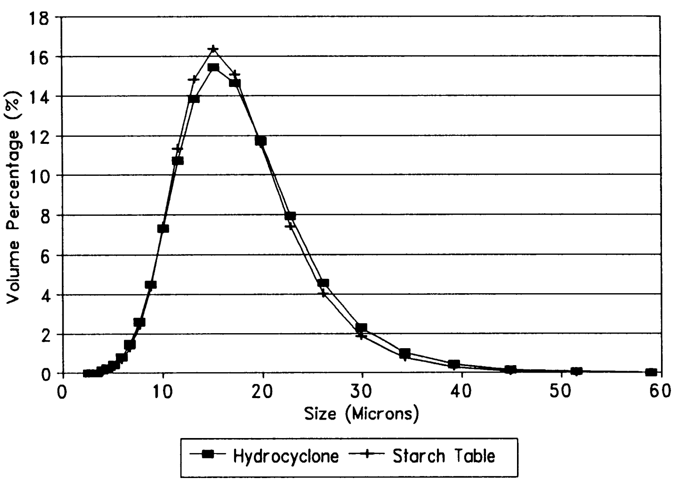

Fig. 2. Particle-size distribution of starch powder obtained by starch tabling and hydrocyclone (Doxie 5) methods for regular dent corn.

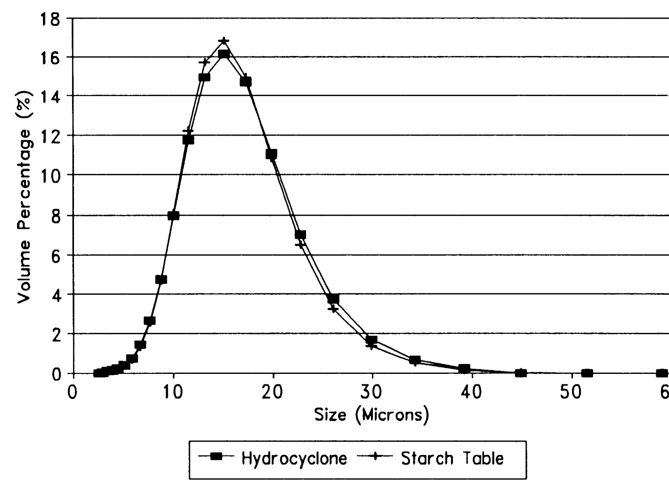

Fig. 3. Particle-size distribution of starch powder obtained by starch tabling and hydrocyclone (Doxie 5) methods for waxy corn.

significantly. The hydrocyclone also eliminates the need for a large floor area to accommodate long starch tables (610 cm long and 10.6 cm wide). Fewer manual operations greatly reduce the potential for operator error in the hydrocyclone procedure. The only critical point in starch-protein separation using the hydrocyclone was setting the inlet pressure to the hydrocyclone. The starch tabling procedure, on the other hand, required decanting water from the mill starch slurry, adjusting the specific gravity of the mill starch slurry, and adjusting the flow rate of the mill starch slurry on the table. Variations in the specific gravity or the flow rate of the mill starch slurry on the starch table can affect the starch yields. The slope of the table was also a critical factor for starch-protein separation. Furthermore, the starch that had settled on the table had to be gently rinsed by the operator, which contributed to operator error. Using a hydrocyclone for starch-protein separation in the laboratory emulates more closely the starch-protein separation process used in industrial operations (i.e., centrifuges and 10-mm hydrocyclones).

Particle-Size Distribution of Starch Powders Obtained by Starch Tabling and Hydrocyclone Procedure

Figure 2 shows the particle-size distribution (averaged for three runs) of starch powders obtained by using starch tabling and the Doxie 5 hydrocyclone for regular dent corn. The volume percentage of small-sized starch particles was higher when using a starch table and the volume percentage of large-sized starch particles was higher when using the hydrocyclone. Because small-sized starch particles end up in the overflow (protein fraction) when starch-protein separation was performed using hydrocyclone, the volume percentage of large-sized starch particles was increased and the volume percentage of the small-sized starch particles was decreased.

For the waxy corn blend, the particle-size distribution using both methods was similar to that obtained for regular dent corn. Figure 3 shows the particle-size distribution (averaged for three runs) of starch powders obtained by using the starch tabling and the Doxie 5 hydrocyclone for the waxy corn.

Comparison to Literature Values

Milling regular dent corn and waxy corn using a Doxie 5 hydrocyclone resulted in higher starch yields and almost the same residual protein content in starch reported in the results of Rubens (1990), who used 10-mm hydrocyclones (TM 1 and TM 3, Dorr-Oliver) for starch-protein separation in a pilot-scale study. On dent corn, Rubens (1990) found a starch yield of 58.8% with a protein content of 0.63%; whereas, the present study had a yield of 62.6% with 0.64% protein. On waxy corn, Rubens (1990) found a starch yield of 55.1% with a protein content of 0.7%; whereas, the present study had a yield of 59.05% with a protein of 0.66%. Steinke and Johnson (1991), who used sedimentation and centrifugation for starch-protein separation, found a starch yield of 58.4% with 0.56% protein, for yellow dent corn. Other laboratory and pilot-scale milling studies (Watson et al 1951, Anderson 1963, Watson 1984, Eckhoff and Tso 1991, Eckhoff et al 1993) used starch tabling for starch-protein separation and reported higher starch yields with lower residual protein content in starch than was found in this study. All these data indicate that the centrifugal separation of starch and protein in the hydrocyclone at laboratory scale is not quite as efficient as the sedimentation method of starch tabling. However, the hydrocyclones are faster, require less space, have higher capacity, and are more analogous to commercial separation techniques.

CONCLUSIONS

A five-pass hydrocyclone system can be used for starch-protein separation instead of starch tabling for 1-kg samples (standard deviation of starch measurement, 0.55%). The tabling procedure gave 3–4% higher starch yields in dent corn; 2–3% higher starch yields in waxy corn; and lower protein contents in starch (0.33 vs. 0.64 for dent; 0.45 vs. 0.65 for waxy) when compared to the hydrocyclone procedure. The advantage of using the hydrocyclone procedure lies in the time required for starch-protein separation, which was reduced by ~75% when compared with the starch tabling procedure. Furthermore, the hydrocyclone procedure required less floor area, considerably reduced the potential for operator errors, and emulated the industrial process at the laboratory scale.

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