FUNCTIONAL CHARACTERISTICS OF AN INTERMEDIATE AMYLOSE STARCH FROM SMOOTH-SEEDED FIELD PEAS COMPARED WITH CORN AND WHEAT STARCHES[1]

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ABSTRACT

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Smooth field peas (*Pisum sativum*) were processed by air classification of pin-milled flour to yield a starch-rich fraction. This starch fraction (34% amylose) was further refined by defibering through screens and water washing to yield a white product containing 0.06% N. This intermediate amylose starch, with granules 20–40 μ in diameter, had a 98% birefringence end point at 70° C. Starches from both smooth-seeded peas and wheat were susceptible to damage by pin milling, whereas the smaller granules of corn starch were less susceptible. Damaging pea starch increased its viscosity during pasting; similarly treated wheat and corn starches had slightly decreased viscosities compared with the undamaged controls. Scanning electron microscopy showed the presence of shattered granules and the exfoliation of the granule surface incurred by pin milling in both pea and wheat starches. The pasting curves of smooth pea starch showed restricted-swelling characteristics similar to those shown by chemically cross-linked starches. Retrogradation of the cooked pastes resulted in a rigid, opaque, friable gel with a firmer texture than corn gels. Syneresis was evident. Pastes of smooth pea starch, gelled in either acidic or basic solutions, demonstrated characteristic differences when compared with corn or wheat pastes. Retorted slurries had higher gel viscosities following retrogradation than did corn starch controls.

In recent years considerable interest has been shown in the use of grain-legume crops in the Prairie regions of Canada. These crops are seen as an agronomically viable alternative to the traditional grains. Such N-fixing crops can both capitalize on potential savings in nitrogenous fertilizer and provide a supply of vegetable protein for animal and human food.

In laboratory and pilot-plant facilities used for this study, techniques have been developed for the production of protein and starch concentrates from field peas and horsebeans by application of pin-milling and air-classification techniques (1,2). Much research effort has been directed toward investigating the potential of field peas because of their agronomic superiority (3) and lack of toxic components when compared with horsebeans (4).

The starchy concentrates are obtained in 50–55% yield by air classification of pea flour and these concentrates can then be further refined to give pure starch in 75 to 85% yield. It is the object of this paper to discuss some of the physical and chemical characteristics of this starch.

MATERIALS AND METHODS

Processing

Smooth field peas (*Pisum sativum* L. var. Trapper) were provided by ProStar Mills Ltd. of Saskatoon, Sask., Canada. Whole or dehulled seeds were pin milled to yield flours of less than 325-mesh particle size in an Alpine Pin Mill

[1]NRCC No. 16280.

Model 250 CW. The flour was then fractionated into protein and starch concentrates using an Alpine Air Classifier Type 132 MP (2). The starch concentrate was mixed with water to yield a 10% suspension, and defibered by filtering first through a 150-μ and then through a 50-μ 300° DSM screen (Dorr-Oliver, Inc., Chicago, IL). The starch was then washed by repeated centrifugation using a Merco Model 9 nozzle-discharge centrifuge, and finally dewatered with a Fletcher basket centrifuge and the cake dried in a forced-air oven.

When it was desired to avoid incurring starch damage, a complete wet process was used. In this process, dehulled field peas were steeped in water containing 0.05% SO_2 for 5 hr, and ground in a Bauer plate mill to yield a coarse slurry. The starch was then defibered and washed as above. The N contents of pea starch prepared by either of these processes were less than 0.06%.

Starches were also prepared on a laboratory scale from several smooth- and wrinkled-pea varieties, and from horsebeans (*Vicia faba equina* L. var. Diana provided by Northern Sales Ltd. of Winnipeg, Manitoba). Flours (40-mesh) were prepared with a Wiley mill and slurried in water. These suspensions were defibered by screening (200 mesh) and the starch was obtained by repeated washing and settling procedures. Starches were dried in a vacuum oven at 60°C/10 hr. Samples of refined corn and wheat starch were kindly provided by the A. E. Staley Co., Decatur, IL.

Microscopy

Structural characteristics of the starches examined in this work were determined with a scanning electron microscope (Cambridge Stereoscan Mark II) using the Hummer gold coating procedure (Technics Co.) for sample preparation. Samples were first dusted onto adhesive tape attached to circular (1.3-cm diameter) aluminum studs prior to coating with approximately 100 Å of gold.

Analytical Procedures

Moisture was determined as described in AACC Approved Method 44-15A (5). Nitrogen content was determined by use of a Hewlett Packard 185B CHN Analyzer. Protein was calculated as %N × 6.25. Iodine affinities of the starches were determined by potentiometric titration (6), with pea amylose absorbing 185 mg iodine/g amylose.

Amylograms were prepared on a Brabender Visco-Amylograph with a 700 cm-g sensitivity cartridge at 75 rpm. The procedure used was according to AACC Approved Method 22-10 (5) with a heating and cooling rate of 1.5°C/min. The slurry pH was adjusted where necessary by addition of 0.1*N* HCl or NaOH as required.

Starch damage was assayed by measuring the percentage of granules susceptible to hydrolysis to glucose by a mixture of α-amylase (Dex-Lo® from Wallerstein Co., Morton Grove, IL) and glucoamylase (Diazyme L-100 from Miles Laboratories Inc., Marschall Division, 1127 Myrtle St., Elkhart, IN). The procedure was a modification of that described by Sandstedt and Mattern (7). Samples of vacuum-oven-dried starch (100 mg) were mixed with 5 ml 0.1*M* acetate buffer pH 4.8 containing 2.5 mg of each enzyme. Tubes were maintained at 50°C and 250-μl aliquots were taken at intervals and assayed for reducing sugar (8). Analysis by linear regression and extrapolation to the ordinate yielded the percentage starch damage data. Starch content was assayed by the dual-enzyme semi-micro method of Banks *et al.* (9).

Retorted-Starch Products

Starch slurries (3% w/w) were sealed in No. 2.5 cans and retorted in an oil bath with shaking at the required temperature for 15 min. Thermocouples were attached to control cans to ensure that suspensions were held at the maximum temperature for 15 min. The pH of the gels was adjusted by use of either 0.2*M* acetate buffer (pH 4.0) or 0.2*M* maleate buffer (pH 6.0). The cans were rapidly cooled by shaking in a cold-water bath. Viscosity of the retorted-starch gels was assayed after 16 hr incubation at 22°C using a Brookfield Synchro-lectric Viscometer Model RVT with the appropriate T-bar and with use of the Helipath Stand. Pasting characteristics were also studied using a pressure Brabender apparatus (courtesy of M. Tessler, National Starch Co., Bridgewater, NJ).

The birefringence end-point temperature was determined as described by Watson (10) using an electrically heated hot stage from a Leitz melting-point apparatus.

RESULTS AND DISCUSSION

The field peas used in this study contained 25.7% protein and 43.7% starch, and were processed by pin milling and air classification to yield a flour containing 78% starch as determined by the dual-enzyme procedure (9). This flour was then further processed by slurrying, filtering, washing, and drying as described in the **METHODS** section to yield an extremely white starch containing less than 0.06% N.

Potentiometric titration experiments indicated small but significant differences between the iodine affinities of the starches from different varieties of smooth field peas obtained by the small-scale laboratory procedure described in the **METHODS** section. These ranged from 5.6 to 7.0% reflecting variations of 30.3 to 37.8% amylose in the starch. The starch used in this study, from the variety Trapper, obtained by the pilot wet- or dry-milling procedures, with undamaged and damaged granules, respectively, was found to contain 34% amylose.

Gelatinization Characteristics

The gelatinization temperature is often cited as a useful characteristic of starch. The birefringence end-point temperature is the temperature at which there is a 98% loss in birefringence when viewed with a microscope equipped with polarizing optics. Some degree of caution has been advised when applying the birefringence end-point temperature too rigorously as an indicator of the starch-gelatinization temperature (11). The polarization crosses of both the grains of smooth field-pea starch and those of a corn starch standard started to decrease in number at 64°C and a 98% loss of birefringence was observed at 70°C. Horsebean starch demonstrated a similar birefringence loss at a slightly lower temperature range of 62–66°C. These effects were observed in starches obtained by either dry- or wet-milling procedures, and occurred irrespective of degree of starch damage. In contrast, compound-starch granules (60.5% amylose) obtained by processing wrinkled-seeded field peas (var. Venus) showed no loss of birefringence nor swelling and bursting of the granules at 98°C. The latter has also been reported by Greenwood and Thomson (12), although these authors noted that smooth field-pea starch had an average gelatinization temperature (the temperature when 50% of the granules were gelatinized) of over 98°C. This is not substantiated by our observations on smooth-pea starch or by those of others (13). Lineback and Ke (14) have reported a gelatinization temperature of 61°–70°C for horsebean starch containing 30% amylose.

Processing and Starch Damage

Isolation of pure starch from legumes is difficult owing to the presence of a highly hydrated fine-fiber fraction. Such fiber is probably derived from the cell walls enclosing the starch granules (13) and was present in starch concentrates prepared from mechanically dehulled seeds. In our laboratory, starch yields from air-classified, pin-milled legume flour were optimized by defibering using a 300° DSM screen (150-μ and 50-μ screens).

The pea-starch slurries were found to have much higher viscosity values when compared with wheat or corn slurries of equivalent solids contents (Table I). In comparison, 50% corn-starch slurries had viscosities of only 2500 centipoise (cP) at 25°C, and 52% wheat slurries had viscosities of 6500 cP at 25°C. In both these cases, 48% slurries had viscosities of 450 and 600 cP, respectively. These

TABLE I
Effect of Concentration on the Viscosity of Pea-Starch Slurries Compared with Corn and Wheat Starches

Concentration Solids %	Starch-Slurry Viscosity Pea cP	Corn cP	Wheat cP
40	1,050	<100	<100
44	20,000	<100	<100
46	92,000	240	360
48	>300,000	450	600
50	...	2,500	1,000
52	...	...	6,500

TABLE II
Effects of Repeated Pin Milling on Starch Damage of Pea, Corn, and Wheat Starches

	Damage/Number of Repeated Passes through Pin Mill			
	0 Passes %	2 Passes %	4 Passes %	6 Passes %
Pea starch	0	21.8	39.6	48.4
Corn starch	0	5.8	8.8	...
Wheat starch	12.1	23.9	32.6	...

processing characteristics are important considerations when designing a scale-up of operations.

During the milling of legume seeds, the starch grains are subjected to considerable shear effects. The degree of damage incurred as a result of this process as applied to pea, corn, and wheat starches is given in Table II. It is apparent that repeated pin milling of pea and wheat starches resulted in substantial granule damage (four passes through the mill resulting in 40% damage in pea starch and 33% damage in wheat starch). The smaller corn-starch granules appeared more resistant to damage by attrition, and four passes through the pin mill only resulted in 8% damage.

These results were substantiated by scanning electron micrographs of these starches. In Fig. 1a, the undamaged-pea starch prepared by the wet-milling process can be compared with the same starch that had been pin milled two times (Fig. 1b), four times (Fig. 1c), and six times (Fig. 1d, higher magnification). Figure 1a demonstrates the heterogeneous population of round to oval grains with shallow-superficial grooves characteristic of most legume starches (15). The grains averaged 20–40 μ in diameter.

These micrographs indicate that attrition grinding resulted in both shattering of the granules as evidenced by the clean-shear lines (Fig. 1b) and also in a superficial exfoliation of the granule surface. This phenomenon is clearly demonstrated in Fig. 1d; enzymatic assay indicated a 48% damage in this sample. In Figs. 1e and 1f, micrographs of corn and wheat starches subjected to four repeated passes through the pin mill are presented. There appeared to be little impact cleavage of the corn-starch granules, but there was superficial pitting and exfoliation apparent. Repeated milling of the wheat starch resulted in a fragmentation of the larger granules together with some superficial exfoliation (Fig. 1f).

Paste Characteristics

Brabender hot-paste viscosity characteristics of the smooth-pea starch are given in Fig. 2. According to Schoch and Maywald (13) these restricted-swelling pastes are classified as Type C starches. Such stabilized hot-paste Brabender viscosities are similar to those of chemically cross-linked starches; possibly a result of formation of intermolecular-hydrogen bonds (16). The wrinkled-pea starch (60.5% amylose) behaved in a manner similar to high-amylose corn starch in terms of its pasting characteristics with no gel development when pasted in 8% w/w suspensions.

The intermediate amylose content of smooth-pea starch results in a greater degree of retrogradation during cooling of the gels. This produced a rigid, irreversible, opaque, friable gel with a firmer texture than comparable corn gels. Syneresis was observed, ranging from a water weep loss of 13 to 15% when gels were pasted at pH values near neutrality to a 5 to 6% weep loss when pasted at pH 3 to 4 and stored at 5°C for 24 hr.

Effects of Starch Damage on Pasting Characteristics

These data are presented in Fig. 3. Repeated milling resulted in a slight overall decrease in viscosity during the heating-cooling cycle with both the wheat and corn starch when compared with the nonpin-milled controls. The effects observed with smooth-pea starch (Fig. 3a) are interesting in that the maximum

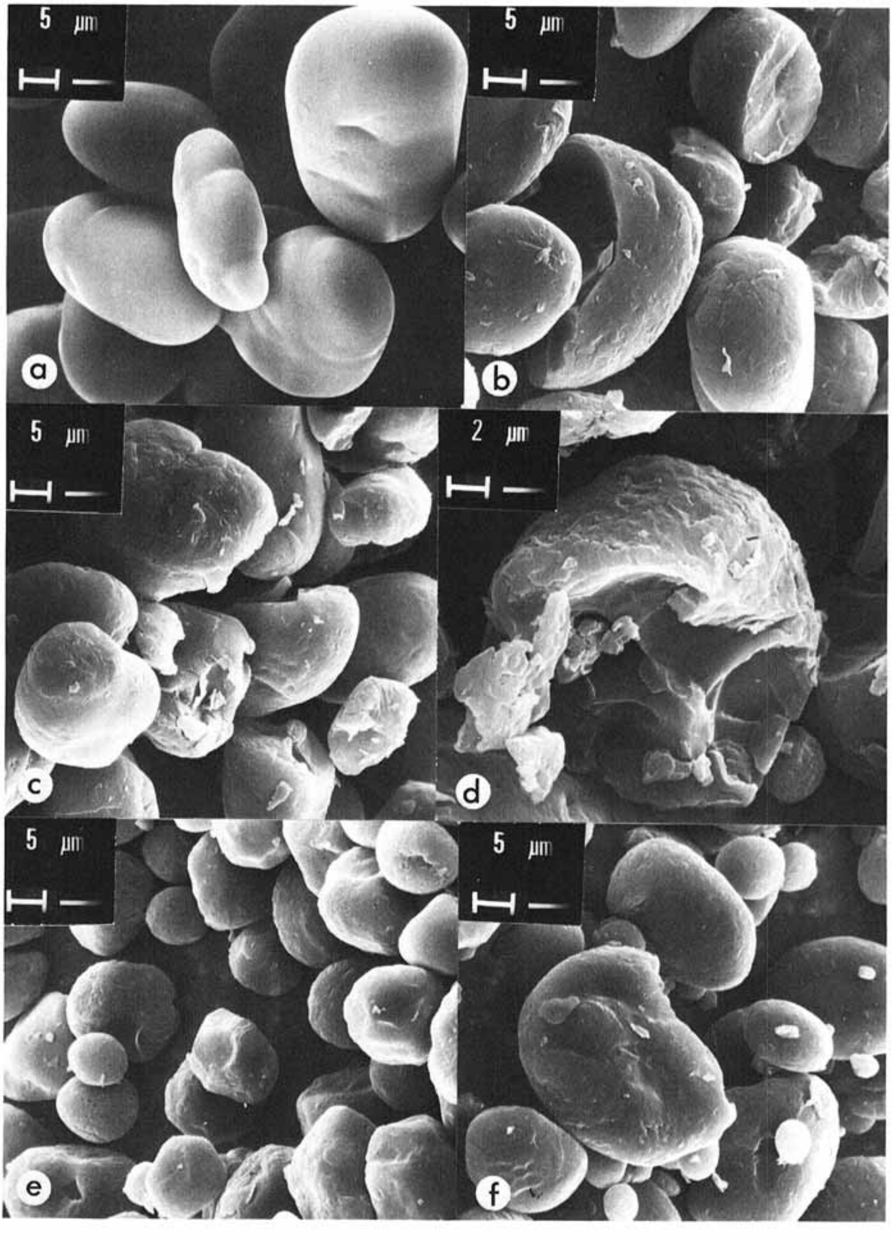

Fig. 1. Scanning electron micrographs of pea, corn, and wheat starches subjected to attrition by pin milling. 1a–d, Pea starch; 1e, corn starch; 1f, wheat starch.

viscosity observed during pasting was increased by almost eight-fold when the starch had 40% damage. Similarly, the degree of set-back was increased by a factor of about three when 40% damaged-pea starch was compared with the zero-damage control.

The induction of improved functional properties in high-amylose starches (50% amylose) by attrition grinding through mechanical rolls has been reported (17). In our studies, high-amylose starch was prepared from wrinkled peas (var. Venus) with 60.5% amylose by the small-scale laboratory wet-milling process. This starch, together with a high-amylose corn starch (58% amylose) was subjected to repeated pin milling (six passes); damage was incurred, but no increase in dispersibility and no viscosity was observed during a Brabender amylogram of 8% slurries.

Effects of Acidic and Basic Conditions during Pasting

As attrition grinding resulted in both increased viscosity at the pea starch pasting peak and increased retrogradation, it appeared of interest to study how this damaged intermediate-amylose starch from field peas would behave under other rigorous conditions. Pasting at low pH values caused degradation of corn and wheat slurries. The effects of this treatment on pea starch are shown in Fig. 4. At acidic pH values up to pH 4, there was a steady increase in viscosity during the

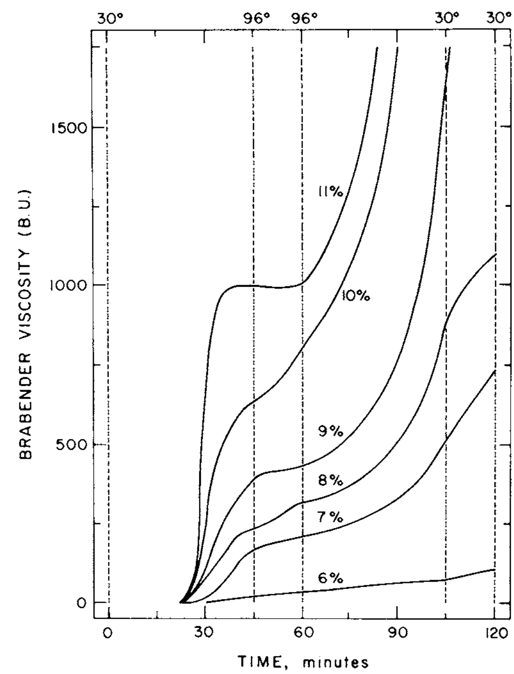

Fig. 2. Brabender pasting curves of pea starch prepared from double-milled air-classified pea flour at the range of concentrations indicated and adjusted to pH 6.5.

pasting program, including a resistance to breakdown under shear when 8% slurries were maintained at 96°C for 15 min. At pH 3, a pasting peak of 620 Brabender units (BU) was observed at 90° C, followed by a decrease in viscosity and then set-back during cooling to 2200 BU.

Pea starch also displayed a marked response to heating in 0.5% w/v sodium hydroxide solution (Fig. 5). Many industrial reactions involve dilute-caustic treatment of starch, *e.g.*, in adhesive formulations for corregated-board production. It is apparent from these data that, when using pea starch in the formulation, decreased levels of starch and/or caustic are required for these applications in order to maintain desired functionalities. Decreased starch content is necessary in these formulations to obtain satisfactory viscosity values and degree of tack, whereas decreased caustic is required to prevent gelation occurring at temperatures below 55°–60° C.

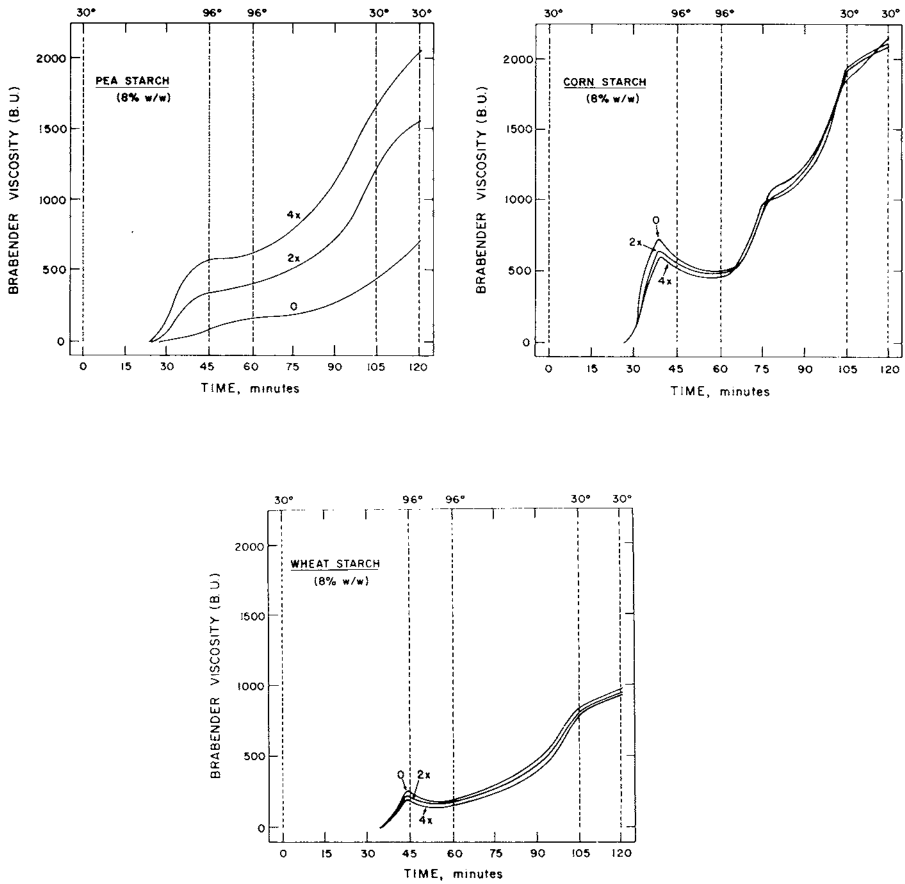

Fig. 3. Effect of pin milling on pasting curves of pea, corn, and wheat starch.

Stability of Pea-Starch Slurries under Retort Conditions

Using a pressurized Brabender apparatus with rapid heating to 140° C of damaged-pea starch slurries (7% w/v), a pasting peak of 320–340 BU was obtained at pH values of 6.0 and 4.8, respectively. Undamaged pea starch (7%

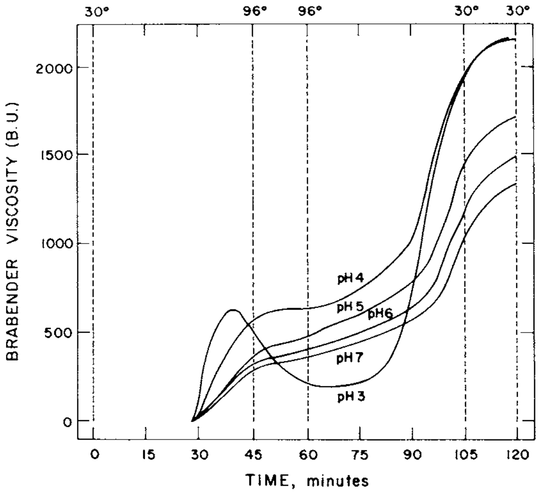

Fig. 4. Effect of acidic pH on pasting curves of pea starch.

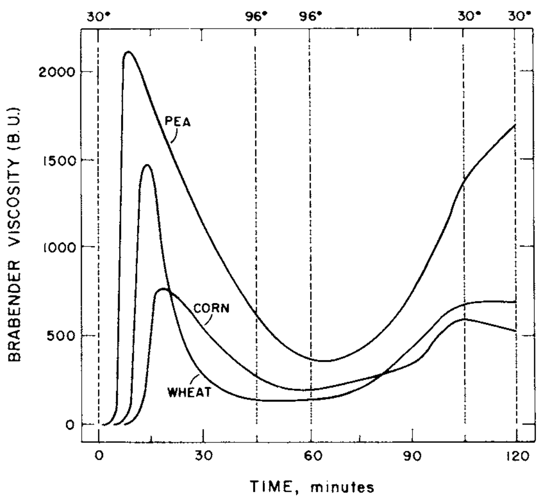

Fig. 5. Effect of dilute caustic (0.5% NaOH) on pasting curves of pea, corn, and wheat starch (8% w/w).

TABLE III
Viscosity of 3% w/w Damaged-Pea Starch Suspensions following Retorting at 95°, 120°, and 130°C for 15-min Intervals

	Gel Viscosity		
	Canning temperature		
	95°C cP	120°C cP	130°C cP
Pea starch			
Pin-milled 2× and water-washed, pH 6.0	4,600	19,300	4,200
Pin-milled 2× and water-washed, pH 4.0	10,600	9,600	500
Pin-milled 2× and air-classified, pH 6.0[a]	600	8,500	6,300
Pin-milled 2× and air-classified, pH 4.0[a]	1,000	4,200	160
Corn starch			
pH 6.0	600	3,400	3,100
pH 4.0	1,000	600	200

[a]The water-washed pea starch contained 0.06% N, air-classified pea-starch flour contained 0.74% N, and the corn starch was a commercial sample containing 0.04% N.

w/v) gave a pasting peak of 350–395 BU when heated in a pressurized Brabender apparatus to 140°C followed by rapid cooling. Much firmer gels were obtained upon set-back of the retorted undamaged starch compared with those pea starch samples prepared by pin milling with consequent granular damage.

Starch slurries, produced from pin-milled flour and containing *ca.* 20% damaged granules, were also sealed in cans and gelled under retort conditions. The viscosities of these 3% w/w gels were determined following retrogradation; the data are presented in Table III. The response of the air-classified pea starch concentrate to retorting conditions is also given. The stability of pea starch gels when held at 120°C for 15 min at both pH 6.0 and pH 4.0 is apparent—a phenomenon more usually associated with chemically cross-linked starches. In this regard, it is of interest to record the observations of Halbrook and Kurtzman (18) who noted that various bean starches had a maximum water uptake at 121°C, considerably higher than that recorded for potato and corn starch. Such properties could be of value for thickening foods requiring high temperatures during processing.

Smooth-pea starches are currently finding applications in commercial markets owing in part to their unique chemical and physical characteristics. Phosphorylated derivatives of pea starch have found applications in the production of pressure-resistant microcapsules used in the manufacture of carbonless paper (19). Plant-scale tests have also demonstrated the value of pea starch in the preparation of corrugated-board adhesives and as clay depressants in potash-mining operations. These developments are contributing to the establishment of a viable field-pea processing operation in the Canadian Prairies.

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