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# Starch characteristics of dry peas (Pisum sativum L.) grown in the USA

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#### ABSTRACT

Starch samples from seven major dry pea (*Pisum sativum* L.) cultivars grown in North Dakota, US was isolated and their physicochemical properties investigated. The objective of the study was to establish the basic foundation of advanced research on physical and chemical modification to improve the functionality of dry pea starches grown in the region. Isolated starch samples were analysed by high performance size exclusion chromatography (HPSEC). Amylose percentages were in the range of 32.2–41.1%. Granules were elliptical or spherical with smooth surfaces based on microscopic analysis. Starch samples had similar gelatinisation transition temperatures and displayed 'C patterns'. The swelling factors of dry pea starches influenced by temperature were determined at 10° intervals between 50 and 90 °C with continuous mixing. The pasting profiles were studied using a rapid visco analyzer (RVA), which exhibited different pasting profiles. This study indicated that dry pea cultivars grown in the region possess starch with different physicochemical characteristics.

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# 1. Introduction

Dry pea (*Pisum Sativum* L.) is a cool-season legume crop, which is grown on over 6.7 million hectares worldwide. The total production worldwide is estimated to be 10.3 million metric tonnes (Agriculture and Agri Food Canada, 2008). Dry pea is marketed as a dry, shelled product for either human or livestock food. Canada, France, China, followed by Russia, India and the United States are the major dry pea producers in the world. Europe, Australia, Canada and the United States raise over 1.8 million hectares and are the major exporters of dry peas. In 2006, there were approximately 374,740 hectares of field peas grown in the United States (USDA Ag Statistics http://www.nass.usda.gov/).

Historically, dry pea was primarily grown in the Palouse region of Washington and Idaho in the US. In the 1990s, North Dakota and Montana began producing dry peas. In 1991, approximately 647 hectares of dry peas were planted in North Dakota; in 2006, this number increased to 247,000 hectares, and accounts for 66% of the US production. The majority (>70%) of the dry pea produced in the United States is exported to India, China and Spain for food and feed processing (McKay, Schatz, & Endres, 2003).

Total carbohydrates of food legumes vary from 24% (winged beans) to 68% (cowpeas). Starch is the most abundant carbohydrate in the seed (22–45%) (Ratnayake, Hoover, Shahidi, Perera, &

Jane, 2001), and is used as an ingredient to modify the texture of food products. Thus, information on starch characteristics (i.e., pasting profiles, thermal behaviours, thickening and gelling properties, swelling factors, etc.) in the starch-water system/food system are important to improve the texture of food products such as frozen foods, extruded snacks, cookies, crackers, sauces, and soups.

Food product texture is significant not only for food processing but also for consumer acceptance. Determination of pea starch properties will make it possible to utilise starch as a suitable ingredient in food products (BeMiller, 2007; Czuchajowska, Otto, Paszczynska, & Baik, 1998). Extensive research has been reported on potato, corn, cereal, and cassava starches because they are used extensively in food and non-food industries. However, there is a demand to investigate pea starch properties in the US food industry primarily for frozen food, pasta, noodle, and bakery products (Personal communication with NPGA, Bismarck, ND).

Previously, physicochemical properties of starches from dry peas grown in Canada have been characterised (Ratnayake et al., 2001). However, there is very limited information available on the physicochemical properties of dry pea starches grown in the US. Therefore, the objective of this research was to investigate the chemical composition, granule morphology, thermal properties, crystallinity, and pasting profiles of starches isolated from seven cultivars of dry pea grown in North Dakota. The outcome of this study will provide basic information for advanced research on physical and chemical modification to improve the functionality



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of dry pea starches, such as modified starch applications in vermicelli noodles; in addition, it will help plant breeders to develop dry pea cultivars with desirable starch functionality.

# 2. Materials and methods

# 2.1. Materials

Seven dry pea cultivars (CDC Mozart, Eclipse, DS Admiral, Miami, Majoret, Cruiser, and Nitouche) were obtained from North Dakota State University (NDSU), Carrington Research Extension Center (CREC) in 2006. CREC is located within a region of North Dakota State that represents a broad diversity of crops. Dry pea varieties were planted on 5 May and harvested by 30 July, 2006. Varieties were grown after a hard red spring wheat rotation. These cultivars represent more than 70% of the dry peas grown in the region (Personal communication with NPGA, Bismarck, ND), thus, indicating the major production potential of the US.

Kits for total starch and starch damage determination were purchased from Megazyme International Ireland Ltd. (Bray, Co. Wicklow, Ireland). The remaining chemicals were purchased from Sigma Chemical Co. (St. Louis, MO, USA).

#### 2.2. Isolation of starch

The pea starch was isolated according to the modified method of Huang et al. (2007). The peas were steeped overnight at a 1:3 (w/v) ratio of peas to a 0.05% sodium bisulphite solution. The peas were then rinsed and milled in a blender with 400 mL deionised water. The ground peas were wet sieved through 50, 100, and 200 mesh Tyler screens. The slurry was centrifuged at  $1509 \times g$  (in 250 mL centrifuge tubes) for 5 min to remove excess water. The starch cake was dispersed in 120 mL of 0.02% NaOH and allowed to settle for 1 h at 4 °C. The starch was centrifuged as before and dispersed in 120 mL deionised water and the pH adjusted to 6.5 with 1 M HCl. The starch was centrifuged again and washed twice with 95% ethanol and once with acetone between each ethanol wash. The starch samples were dried overnight at 30 °C.

## 2.3. Chemical composition of the isolated starches

The determination of moisture, protein, total starch, and starch damage for the pea samples were carried out using ACCC (2000) approved methods. The moisture was determined using the approved method 44-15A, and the crude protein was determined using the nitrogen combustion approved method 46-30 (AACC International, 2000). The total starch assay kit was used to determine the total starch content (% dry weight basis, dwb) of each of the samples (Approved Method 76-13, AACC International 2004). The starch damage assay kit was used to determine the starch damage (% dry weight basis) of each of the flours (Approved Method 76-31, AACC International 2004).

# 2.4. Starch molecular weight distribution

The starch samples were treated with KOH and urea before high performance size exclusion chromatography (HPSEC) analysis as described earlier by Grant, Ostenson, and Rayas-Duarte (2002). The starches ( $\approx$ 20 mg) were solubilised by adding 4.5 mL of 1.0 M KOH and 0.5 mL of 6.0 M urea and heating at 100 °C under nitrogen for  $\approx$ 90 min. After this heating, 1 mL of the sample was neutralised with 1.0 M HCl and filtered through a hydrophilic nylon syringe filter before analysis. A Waters ultrahydrogel Linear 6–13 µm, 7.8-mm × 300-mm column with an ultrahydrogel guard column (Waters, Milford, MA) were used to analyse starches. The

samples were run using a Agilent 1100 series high-performance liquid chromatograph (Agilent Technologies, Wilmington, DE), equipped with an auto sampler. An Agilent refractive index detector and PC with ChemStation (HP ChemSation for LC Rev. A.04.01) were used for control and integration. The samples were analysed at 40 °C with filtered deionised, distilled water as the mobile phase. The flow rate was 0.4 mL/min and injection volume was 10 µl.

#### 2.5. Chain length distribution of starch

Twenty milligrams of each type of starch were dispersed in 500 µl of 90% dimethyl sulphoxide (DMSO) and heated in a boiling-water bath to ensure a complete dispersion. From the dispersion, 140 µl was aliquoted, and a sodium acetate buffer (0.02 M, 1000 µl, 50 °C, PH 4.75) was added. The mixture was heated again in a boiling-water bath for 10 min and cooled to 37 °C in a shaking water bath. Isoamvlase solution (Megazyme, 10 U/mL, 30 ul in acetate buffer) was added to each mixture. The mixture was incubated for 24 h at 37 °C with shaking and then boiled for 10 min to denature the enzyme. For each mixture after debranching, moisture was removed using a SpeedVac concentrator (Savant) with simultaneous heating and vacuuming, and the volume of each sample was adjusted to 1 mL with 90% DMSO. After vortex and centrifugation to remove insolubles, a 20 µl aliquot was injected into to the HPSEC system. The HPSEC system contained two connected Zorbax gel PSM 60-S columns ( $6.2 \times 250$  mm, Agilent Tech., Santa Clara, CA) and a flow rate of 0.5 mL/min, with DMSO as the mobile phase. The elution was monitored by a Waters 2414 refractive index (RI) detector (Waters, MA). Glucose, maltose, malto-pentaose (Degree of polymerisation 5, DP5), and pullulan with 5900, 11,800, 22,800, 47,300, 112,000, and 212,000 Da (Polymer Laboratories, Amherst, MA) were used for column calibration.

### 2.6. Granule morphology

Starch samples were mounted on aluminium mounts using colloidal silver or carbon adhesive tabs and coated with gold using a Balzers SCD 030 sputter coater (BAL-TEC RMC, Tucson, AZ). Images were obtained using a JEOL JSM-6300 Scanning Electron Microscope (JEOL USA, Peabody, MA) while using an accelerating voltage of 15 keV.

# 2.7. Swelling factor (SF)

The swelling factor for the isolated starch samples was determined using a modified method described by Huang et al. (2007). Starch slurries of each sample were made to contain 0.20 g starch and 10 mL deionised water. The starch slurries were heated and stirred using rapid visco analyzer 4 (RVA4). The slurries were equilibrated at 25 °C for 30 min before heating at 50, 60, 70, 80, or 90 °C for 30 min, and then cooled to 25 °C before centrifuging at 1000×g for 15 min after the slurries were transferred into centrifuge tubes. Different starch slurry samples were used for each temperature treatment. The supernatant was removed and the swelling factor (SF) was calculated as; SF (mL/g dry starch) = (10 mL – supernatant volume)/0.2 g starch.

#### 2.8. Differential scanning calorimetry (DSC)

The thermal analyses of starch samples were performed in triplicate with a Perkin–Elmer DSC7 instrument (Norwalk, CT) using the method described earlier (White, Abbas, Pollak, & Johnson, 1990). The onset of gelatinisation ( $T_o$ ), the temperature at peak ( $T_p$ ), the temperature at the completion of gelatinisation ( $T_c$ ), and the enthalpy of gelatinisation ( $\Delta H$ ) were obtained using the data processing software supplied with the DSC instrument.

#### Table 1

Chemical components of ground pea and isolated dry pea starches.

Cultivar name	Chemical analysis <sup>a,b</sup>						
	TS (dwb)	SD (dwb)	TP (as is)	HMW-AP (%)	LMW-AP (%)	T-AP (%)	AMY (%)
DS Admiral (yellow pea)	41.4ab ± 0.54	1.77c ± 0.01	1.11b ± 0.05	34.8a ± 0.59	29.5c ± 0.35	64.3bc ± 0.94	35.7ab ± 0.94
Cruiser (green pea)	40.9ab ± 0.60	$1.67b \pm 0.01$	$0.74a \pm 0.04$	44.1c ± 1.17	23.7ab ± 1.11	67.8c ± 0.06	32.2a ± 0.06
Eclipse (yellow pea)	42.1abc ± 0.31	1.56a ± 0.05	$1.06b \pm 0.01$	37.4ab ± 0.68	21.5a ± 1.93	58.9a ± 1.83	41.1c ± 1.83
Majoret (green pea)	40.5a ± 1.45	1.58a ± 0.03	3.31c ± 0.07	42.5c ± 0.81	22.0ab ± 1.68	64.5bc ± 2.35	35.5ab ± 2.35
Miami (yellow pea)	44.4d ± 0.06	1.80c ± 0.03	0.55a ± 0.06	39.9bc ± 1.28	24.0ab ± 1.93	63.9bc ± 0.64	36.1ab ± 0.64
CDC Mozart (yellow pea)	42.5bc ± 0.07	1.61ab ± 0.04	$1.03b \pm 0.05$	37.6ab ± 2.09	24.7b ± 0.51	62.3ab ± 1.58	37.7bc ± 1.58
Nitouche (green pea)	43.2cd ± 0.26	1.54a ± 0.04	$1.14b \pm 0.21$	40.9bc ± 4.70	24.1ab ± 0.88	65.0bc ± 3.82	35.0ab ± 3.82

<sup>a</sup> Values not sharing a common letter are significantly different ( $P \leq 0.05$ ).

<sup>b</sup> TS = total starch in ground dry pea, SD = starch damage in ground dry pea, TP = total protein in isolated pea starch, HMW-AP = high molecular weight amylopectin, LMW-AP = low molecular weight amylopectin, T-AP = total amylopectin, AMY = amylose, dwb = dry weight basis.

#### 2.9. X-ray powder diffraction

The degree of crystallinity was investigated as described before (Chakraborty et al., 2004). The [%] crystallinity of the starch samples was defined by the intensity ratio of the diffraction peaks and of the sum of all measured intensity. Background intensity is subtracted from the total intensity. Degree of crystallinity was calculated using the following equation:

Crystallinity [%] = 
$$100 \times \sum I_{\text{net.}} / \left( \sum I_{\text{tot.}} - \sum_{\text{cons. bgr.}} \right)$$

*Xpert High Score Plus* (PANalytical, Almelo, Netherlands), version 2.2b software was used to perform calculations.

#### 2.10. Pasting properties

The pasting profiles were analysed using a Newport Scientific rapid visco analyzer 4 (RVA). The extracted starch samples were run using a method defined by Huang et al. (2007). Starch slurries were prepared to contain 6% (w/w) starch with a total weight of 28 g. The starch slurries were held at 30 °C for 1 min before heating at a rate of 15 °C/min to 90 °C for 5 min, and then cooled at a rate of 15 °C/min to 30 °C and held for 7 min.

### 2.11. Statistical analysis

All analyses were replicated ( $n \ge 2$ ) and mean values and standard deviation values were reported. The analyses of variance (AN-OVA) mean values were determined by Duncan's Multiple Range Test (P < 0.05) with the SPSS 15.0 Statistical Software Program.

#### 3. Results and discussion

# 3.1. Starch composition of dry pea

Total starch and starch damage results are given in Table 1. The cultivars showed differences in terms of total starch and starch damage (P < 0.05). Miami and Nitouche had the highest starch content, 44.7% and 43.5%, respectively. Majoret had the lowest total starch with 40.8%. All pea cultivars were obtained from the same location, thus genotypic background may be considered to be a factor of the differences. Starch damage scores were in a range of 1.54–1.80%. Starch damage variation might be associated with the differences of seed hardness. The isolation of starch from pea was reported to be a difficult process since insoluble flocculent proteins and fine fibre found in pea decrease sedimentation and co-settle with starch. Starch with higher protein content can have a darker colour (Ratnayake, Hoover, & Warkentin, 2002; Tyler, Youngs, & Sosulski, 1981). Protein was also detected in starch

samples in our study at a level of 1–2% excluding Majoret, which had 3% protein (Table 1).

#### 3.2. Starch molecular weight and chain length distribution

Isolated pea starches were analysed by HPSEC after solubilisation by KOH, urea and heat treatment. Chromatograms are given in Fig. 1 (top) and integration values for the high molecular weight amylopectin (HMW-AP), low molecular weight amylopectin (LMW-AP), total amylopectin (T-AP) and amylose (AMY) are listed in Table 1. The assignment of the peaks was performed as described earlier (Grant et al., 2002). HMW-AP ratios ranged from 34.8% to 44.1%. Cruiser had the highest amount of T-AP (67.8%) and HMW-AP (44.1%). Eclipse had the lowest amount of LMW-AP (21.5%) and T-AP (58.9%). AMY% ranged from 32.2% to 41.1%.

The chain length distributions of starches are shown in Fig. 1 (bottom). In general, the chromatograms of individual starches shared a similar chain length distribution pattern. The amylose section appeared in the range of over DP70. Three chain populations could be identified from the amylopectin section. Population (I) had a modal length around DP40, which originates mostly from the inter-cluster (B2) chains. Populations (II) and (III) had modal length around DP12, which corresponded to short (intra-cluster) B chains and A chains.

#### 3.3. Starch granule morphology

Analysis of the granule morphology of the extracted starches was performed using SEM (data not shown). Most of the granules were oval, even though round, spherical, elliptical, and irregularly shaped granules were also observed. Surfaces of starch granules were smooth and displayed no fissures or compound granules. Granule length and width ( $\mu$ m) data are given in Table 2. The length of the granules ranged from 26.8  $\mu$ m to 30.7  $\mu$ m. Nitouche showed the largest variation in terms of granule length. Variation in granule width was also detected in all the starch samples. Granules from Majoret had the greatest (27.0  $\mu$ m) width and Nitouche had the least (16.6  $\mu$ m) width.

### 3.4. Gelatinisation characteristics

DSC gelatinisation temperatures and enthalpies ( $T_o$ ,  $T_p$ ,  $T_c$ ,  $T_c-T_o$ , and  $\Delta H$ ) of the isolated starches are listed in Table 3. Starch gelatinisation is an order to disorder phase transition in which intermolecular hydrogen bonds of starch molecules break down in the presence of sufficient water and elevated temperature. This allows the glucan chains to absorb more water. Incorporation of water molecules through hydrogen bonding increases the randomness of glucan chain conformation and decreases the number and size of crystalline regions. Heating immobilises the chain segments in



**Fig. 1.** (Top) High performance size exclusion chromatography (HPSEC) analysis of isolated dry pea starches. Abbreviations; HMW-AP: high molecular weight amylopectin, LMW: low molecular weight amylopectin. (Bottom) Chain length distribution of starches determined using HPSEC analysis of debranched product of starches. The dashed line is used to differentiate the linear chain populations originated from amylose (left) and amylopectin (right). Three chain populations of amylopectin (I), (II), and (III) are labelled.

crystalline regions and allows water absorption, which leads to the reduction of the ordered structure (e.g., double helices and crystallites) and amylose leaching. The gelatinisation process is associated with starch source and affected by starch fine structure (BeMiller, 2007). During starch gelatinisation, transformation from crystallites to an amorphous state starts from the hilum area (Ratnayake et al., 2002) and moves rapidly to the B polymorphs, which are mostly present in the central part of the granule. Following this process, the central part of the granule begins to swell. At this stage, the

# Table 2

Granule size distribution and degree of crystallinity of isolated dry pea starches.

Cultivar name	Starch granule	Starch granule sizes				
	Length (µm)	Length (µm)				
	Mean	Range	Mean	Range		
DS Admiral	29.8	40.8-1.7	18.00	19.2-15.0	8.5	
Cruiser	28.2	35.0-2.5	16.6	19.2-13.3	8.7	
Eclipse	26.7	38.3-4.2	17.8	27.5-11.7	8.7	
Majoret	27.0	35.0-8.3	18.3	23.3-9.2	7.8	
Miami	29.4	34.2-17.5	20.0	35.0-13.3	8.7	
CDC Mozart	29.4	37.5-20.0	18.2	23.3-10.0	8.0	
Nitouche	30.7	41.7-19.2	16.6	17.5–14.2	7.9	

<sup>a</sup> Calculated as described in materials and methods.

Table 3           Thermal properties of isolated dry pea starches determined by differential scanning calorimetry (DSC).				
Cultivar name	Thermal properties <sup>a,b</sup>			

Thermal properties <sup>a,D</sup>						
<i>T</i> <sub>o</sub> (°C)	$T_{\rm p}$ (°C)	<i>T</i> <sub>c</sub> (°C)	$T_{\rm c}$ – $T_{\rm o}$ (°C)	$\Delta H$ (J/g)		
62.9ab ± 0.20	69.1a ± 0.12	74.9a ± 0.17	12.0ab ± 0.10	12.4ab ± 1.16		
63.8b ± 2.28	69.9b ± 0.76	75.1a ± 0.44	11.3a ± 1.85	12.4ab ± 0.78		
63.3ab ± 0.45	68.6a ± 0.17	74.7a ± 0.42	11.3a ± 0.30	10.9a ± 0.72		
62.5ab ± 0.17	68.7a ± 0.20	74.9a ± 0.80	12.4ab ± 0.95	10.7a ± 0.87		
62.5ab ± 0.46	69.0a ± 0.25	74.6a ± 0.30	12.1ab ± 0.76	12.3ab ± 1.05		
61.8ab ± 0.75	68.4a ± 0.38	74.4a ± 0.75	12.6ab ± 0.10	10.9ab ± 1.15		
61.7a ± 0.15	68.9a ± 0.06	75.0a ± 0.53	13.3b ± 0.49	13.3b ± 1.80		
	$\frac{\text{Thermal properties}^{\text{a,b}}}{T_{\text{o}} (^{\circ}\text{C})}$ 62.9ab ± 0.20 63.8b ± 2.28 63.3ab ± 0.45 62.5ab ± 0.45 62.5ab ± 0.17 62.5ab ± 0.46 61.8ab ± 0.75 61.7a ± 0.15	Thermal properties <sup>a,b</sup> $T_o$ (°C) $T_p$ (°C)           62.9ab ± 0.20         69.1a ± 0.12           63.8b ± 2.28         69.9b ± 0.76           63.3ab ± 0.45         68.6a ± 0.17           62.5ab ± 0.17         68.7a ± 0.20           62.5ab ± 0.46         69.0a ± 0.25           61.8ab ± 0.75         68.4a ± 0.38           61.7a ± 0.15         68.9a ± 0.06	$\begin{tabular}{ c c c c } \hline Thermal properties^{a,b} \\ \hline \hline T_{o} (^{\circ}C) & T_{p} (^{\circ}C) & T_{c} (^{\circ}C) \\ \hline $	Thermal properties <sup>a,b</sup> $T_0$ (°C) $T_p$ (°C) $T_c$ (°C) $T_c-T_o$ (°C)62.9ab ± 0.2069.1a ± 0.1274.9a ± 0.1712.0ab ± 0.1063.8b ± 2.2869.9b ± 0.7675.1a ± 0.4411.3a ± 1.8563.3ab ± 0.4568.6a ± 0.1774.7a ± 0.4211.3a ± 0.3062.5ab ± 0.1768.7a ± 0.2074.9a ± 0.8012.4ab ± 0.9562.5ab ± 0.4669.0a ± 0.2574.6a ± 0.3012.1ab ± 0.7661.8ab ± 0.7568.4a ± 0.3874.4a ± 0.7512.6ab ± 0.1061.7a ± 0.1568.9a ± 0.0675.0a ± 0.5313.3b ± 0.49		

<sup>a</sup> Values not sharing a common letter are significantly different ( $P \leq 0.05$ ).

<sup>b</sup>  $T_{o}$ ,  $T_{p}$ ,  $T_{c}$  = gelatinisation onset, peak and conclusion temperatures;  $T_{c}$ - $T_{o}$  = gelatinisation temperature range,  $\Delta H$  = gelatinisation enthalpy.



Fig. 2. Swelling factor analysis of isolated dry pea starches.

crystallites of the A polymorphs, which are arranged in the outer part of the granule, are not disrupted. At higher temperatures, this region is also disrupted and gelatinisation is completed (Ratnayake et al., 2002). Smooth pea starches with a higher content of the B polymorph, which is packed with higher energy capacity than A polymorph, showed higher  $\Delta H$ . On the other hand, smooth pea starch, which has a higher content of A polymorphs, was shown to have higher gelatinisation temperatures (Bogracheva, Morris, Ring, & Hedley, 1998; Davydova, Leontev, Genin, Sasov, & Bogracheva, 1995; Ratnayake et al., 2002). Nitouche indicated the highest  $\Delta H$  (13.3 J/g), whereas Majoret had the lowest value (10.7 J/g).  $T_{\rm o}$ ,  $T_{\rm p}$ ,  $T_{\rm c}$ , and  $T_{\rm c}$ – $T_{\rm o}$  values were between 63.8 and 61.7, 69.9 and 68.4, 75.1 and 74.4 and 13.3 and 10.8, respectively. These values were in the range of previously reported pea starch gelatinisation temperatures (Hoover & Ratnayake, 2002; Huang et al., 2007; Ratnayake et al., 2001).

## 3.5. X-ray diffraction

X-ray diffraction has been widely used to determine the degree of crystallinity of starch granules. Usually, legumes display a 'C' type diffraction pattern. C type is an intermediate pattern between A type (cereal starches) and B type (tuber starches) (BeMiller, 2007). Strong peaks at 15.5 Å (characteristics of B type), 17.4 Å (characteristic of A type) and 23.4 Å (characteristic of A type) were detected in X-ray diffraction. Therefore, all dry pea starches exhibited C type diffraction patterns (data not shown). The degree of crystallinity was calculated (Table 2). Miami, Eclipse, and Cruiser varieties displayed a relatively higher degree of crystallinity (8.7%), which correlates with the relatively higher  $T_{o}$ ,  $T_{p}$ ,  $T_{c}$ , and  $\Delta H/T$ -AP values. The degree of crystallinity values in our study was lower than those previously reported values (Ratnayake et al., 2002).

#### Table 4

Pasting characteristics of isolated dry pea starches determined by rapid visco analyzer (RVA).

Cultivar name	RVA Pasting charac	RVA Pasting characteristics <sup>a,b</sup>							
	PV (RVU)	BKD (RVU)	STB (RVU)	HPV (RVU)	CPV (RVU)	PT (min)			
DS Admiral	42.8c ± 0.70	3.5b ± 0.30	28.9ab ± 0.23	39.3d ± 1.00	68.2bc ± 1.24	6.9a ± 0.05			
Cruiser	40.8c ± 0.30	3.2ab ± 0.23	29.1ab ± 1.06	37.6cd ± 0.53	66.7bc ± 0.53	6.9a ± 0.07			
Eclipse	35.4b ± 3.66	3.3ab ± 0.35	25.9a ± 4.48	$32.1b \pm 4.01$	58.0ab ± 8.49	7.0a ± 0.02			
Majoret	40.2bc ± 1.18	3.5b ± 0.18	35.2c ± 1.18	36.7bcd ± 1.36	71.9c ± 2.54	7.0a ± 0.02			
Miami	43.8c ± 3.13	3.1ab ± 0.35	41.6d ± 3.47	40.7d ± 2.77	82.3d ± 6.24	6.9a ± 0			
CDC Mozart	28.0a ± 1.82	$2.8a \pm 0.11$	24.6a ± 0.47	25.2a ± 1.94	49.8a ± 1.47	6.8a ± 0.02			
Nitouche	35.4b ± 0	2.8a ± 0	32.9bc ± 0.29	32.7bc ± 0	65.5bc ± 0.30	$6.8a \pm 0.14$			

<sup>a</sup> Values not sharing a common letter are significantly different ( $P \leq 0.05$ ).

<sup>b</sup> RVA = rapid visco analyzer, PV = peak viscosity, BKD = breakdown, STB = setback, HPV = hot paste viscosity, CPV = cold paste viscosity, PT = peak time.

#### 3.6. Swelling factor (SF)

When starch is heated in the presence of water, the crystalline structure is transformed to an amorphous structure. Exposed hydroxyl groups of amylose and amylopectin form hydrogen bonds with water molecules. This interaction triggers granule swelling. On the other hand, the interactions among glucan chains retain the structural association originated from the granular structure. The equilibrium between granule swelling and retaining governs the swelling factor. A number of factors may affect such equilibrium, which include the ratio and fine structure of amylose and amylopectin, granule-bound protein and lipid compounds, and thermal treatments. The swelling factor of starches influenced by temperature was determined at 10° intervals between 50 and 90 °C with continuous mixing (Fig 2). The rapid increase in the swelling factor was detected between 60 and 80 °C. Although the swelling factor scores of the starches were increased by elevated temperatures, the rate in increase was different for each variety. Majoret had the highest and Cruiser had the lowest swelling factor at 50 °C. At 60 °C, Nitouche had the highest swelling factor followed by Majoret. Again, Cruiser and CDC Mozart had the lowest swelling factor at this temperature. Starch samples had similar swelling factors at 70 °C. Even though Majoret had the highest swelling at 50 °C, it showed the lowest swelling factor at 80 °C. Usually, cultivars that had a relatively higher swelling factor at lower temperatures, exhibited lower swelling factors at higher temperatures. In addition to the interactions among starch molecules, amylose-lipid complexes were shown to limit swelling and solubilisation (Hoover & Hadziyev, 1981). Extensive interaction between closely packed amylose chains and/or higher magnitude of amylose-lipid complexes could contribute to the lower swelling factor of some dry pea starches (Ratnayake et al., 2002), which needs to be further investigated.

#### 3.7. Pasting characteristics

The pasting characteristics of the starches are summarised in Table 4. Even though the overall shape of the RVA curves of starch samples were similar, the differences in peak viscosity (PV), breakdown (BKD), setback (STB), hot paste viscosity (HPV), and cold paste viscosity (CPV) were detected (P < 0.05). Overall pasting profile was similar to other legume starches (Hoover & Sosulski, 1991). All starch samples had very similar peak time (PT). PV was highest for Miami (43.8 RVU) and lowest for CDC Mozart (28.0 RVU). PV values ranged from 35.4 RVU to 42.8 RVU for the other varieties. BKD values were between 2.8 RVU and 3.5 RVU. There was again considerable variation in STB. Miami had the highest STB value (41.6 RVU) and CDC Mozart had the lowest (24.6 RVU). When HPV and CPV values were analysed, again Miami showed the highest values, 40.7 RVU and 82.3 RVU, respectively.

The effect of genotypic differences in the pasting profile of dry pea starches was reported before (Davydova et al., 1995; Ratnayake et al., 2001). In previous studies, pasting properties were mostly analysed using Brabender Visco Amylograph (BVA) in which dry pea starches showed the absence of peak viscosity, increasing viscosity during the holding period (at 95 °C), and a high setback. In addition, the pasting profile of dry pea starches might be influenced by non-starch components in starch samples.

# 4. Conclusions

Starch is the main carbohydrate reserve found in plants. It is a major source of nutrition for humans and animals, and an important raw material for industry. The functional properties of starch determine its applications in industry. One of the important functional properties is pasting, which is the formation of a high viscosity solution after heating of starch-water suspensions. This characteristic of starch is utilised in various foods as well as in non-food applications such as adhesives. Another important functional characteristic is the ability to form gels, which is also used in different foods and in non-food applications such as thermoplastics. Additionally, functional properties of starch are determined by starch granular structure. Starch granules have semicrystalline structure and their functional properties are, therefore, dependent on the structure and proportion of their crystalline and amorphous regions.

In this study, starch properties from seven dry pea cultivars planted in the US were analysed. All dry pea samples were collected from the same location during the same harvesting period. Thus, we believe that the differences in starch properties were considered to originate from the genotypic differences. Previously, a series of near-isogenic lines have been developed for mutations induced and identified at six independent loci controlling steps in starch biosynthesis pea seeds (Hedley, Bogracheva, & Wang, 2002). Starches produced by these pea lines were characterised in terms of the morphology, structure and properties of the starch granules (Bogracheva et al., 1999; Ridout, Parker, Hedley, Bogracheva, & Morris, 2006). Based on these studies, r or rug5 loci encode a starch branching enzyme and a soluble starch synthase in pea. The shape of starch granules from lines containing mutations at either the *r* or *rug5* loci were significantly different from granules found in the non-mutant parental line. Mutations at each of the six loci were investigated using X-ray diffraction, NMR, DSC, polarised light microscopy, and atomic force microscopy (Ridout et al., 2006). The results from these studies indicated that relatively small but important changes were determined in the percentage of crystalline material within the granules. Large differences were found between the mutants in the distribution of A- and B-type crystallites within the granules, ranging from about 60% A-type in rug3 mutants to 100% B-type in r mutant lines. The studies proposed that the pea lines are a very useful resource to answer essential questions on the genetic control of starch granule structure and function (Hedley et al., 2002).

The varieties in this study displayed differences in their starch content and percentages of HMW-AP, LMW-AP, T-AP, and AMY. DSC thermal properties, granule morphology, X-ray diffraction pattern, and pasting profiles were also studied. Miami, Eclipse, and Cruiser seem to have similar crystallinity and thermal properties. AMY percentages ranged from 32.2% to 41.1%. The ratio of LMW-AP was also determined for each cultivar. We observe that the pasting curve of native pea starch is typical of those of other legume starches. Based on the pasting profile analysis, Miami had the highest PV, BKD, STB, HPV, and CPV values. Previous studies showed that the pasting properties of pea starches are significantly influenced by genetic effects (depending on the variety or cultivar) (Davydova et al., 1995; Ratnayake et al., 2001). The molecular characteristics of the starch components could explain the pasting behaviour of field pea starches.

The results of this study will provide the basic knowledge on the starch structure of dry pea starches, and it will help plant breeders to develop dry pea cultivars with the desired starch functionalities.

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