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Some properties of seeds and starches separated from different Indian pea cultivars

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Abstract

The changes in seed hardness and moisture content during storage of seeds from different pea cultivars were studied. The seed hardness values of all pea cultivars differed significantly and NDVP-12 seeds showed the highest seed hardness. Moisture contents of seeds from different pea cultivars decreased and seed hardness increased during storage. Starches were also isolated from different pea cultivars and investigated for morphological and thermal properties. The starches separated from the different pea cultivars had a granule size ranging between 31.81 and 2.72μm while the structure of starch granules varied from simple to complex. The amylose content in pea starches varied between 26.9 and 61.5%. MA-6 pea starch showed a greater proportion of large-size granules while the other pea cultivars had small granules with complicated structures. The transition temperatures, enthalpies of gelatinization (ΔH_{gel}), peak height indices (PHI) and range (R) of starches separated from different pea cultivars were determined using differential scanning calorimetery. The ΔH_{gel} and PHI were highest for MA-6 pea starches and lowest for NDVP-12 pea starch. The value of R was highest for NDVP-12 pea starch but lowest for MA-6 pea starch. The turbidities of gelatinized aqueous starch suspensions, from all pea cultivars, increased with increase in storage period; however, increase was highest in MA-6 pea starch.

Keywords: Pea; Morphological; Thermal; Amylose content; Turbidity; Starch

1. Introduction

Legumes are rich in proteins and contain less starch, than cereals or tubers. Starch constitutes about 45–65% of the composition of edible legumes. Starch constitutes most of the dry matter, accumulating in the harvested organs of the crop plants, and is therefore not only a primary source of calories in the human diet. It can also be regarded as a renewable resource that may be utilized in many industrial applications. Out of 16,000–19,000 species, only 12 species are used widely by the food industry. These include common beans, field peas, chickpea, cowpeas, greengram, blackgram, lentils, pigeonpea and the two oilseeds, soybeans and groundnuts (Deshpande & Damodaran, 1990). Legumes are also used for green manuring, as forage for cattle and to improve the fertility of the soil. Even though starch is almost solely composed of glucose, evolution has generated diversity to such an extent that it is possible to conclude that starches derived from any given plant species are unique. Starches from legumes have been reported to be viscous, indicating high resistance to swelling and rupture earlier, so they can be used as ingredients for various industrial applications (Lineback & Ke, 1975).

Legumes are popular in the human diet for their protein contents. However, detailed information on the structure, gelatinization and retrogradation properties of legume starches is lacking. Studies of structure and phase transition of legume starches are important as these have a profound influence on texture, appearance, water-holding capacity and enzyme digestibility of processed starch foodstuff (Biliaderis, 1991). In order to understand and predict legume starch function, it is important to establish structure property relationships. The development of efficient processing techniques for separating proteins and starch components from legume seeds has opened new paths for further studies of legume starch components from leguminoseae crops. Biliaderis, Grant, and Vose (1979) have explained the pasting properties of starches on basis of their molecular characteristics. Pea starch forms the most abundant carbohydrate (22–45%)

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in seeds (Hoover & Sosulski, 1991; Ratnayake, Hoover, Shahidi, Perera, & Jane, 2001). Pea starches are important functional ingredients in high temperature extrusion processes (Gujska, Reinhard, & Khan, 1994; Otto, Baik, & Czuchajowska, 1997b). Pea starches are of interest because of their relatively high amylose contents (Schoch & Maywald, 1968). Pea starches have been introduced as new food ingredients widely used in the processed meat industry, in canned meats, cooked sausages and patties (Beck & Kevin, 1995; Comer & Fry, 1978; Otto et al., 1997b). In this paper we have studied some properties of pea seeds and pea starches of four different Indian pea cultivars.

2. Material and methods

2.1. Materials

Four pea cultivars (c.v.) i.e. MA-6, VL-7, Arkel, NDVP-12 were procured from Punjab Agricultural University, Ludhiana, India from the 2001 harvest.

2.2. Seed hardness

About 20 seeds of uniform size of the different pea cultivars were stored in a refrigerator at 4 °C and, after every seven days, about five seeds of uniform size were selected for the hardness test. An Instron Universal Machine (Model-4464, Instron, Buckinghamshire, England) was used to measure the seed hardness for different pea cultivars at a crosshead speed of 50 mm/min.

2.3. Starch isolation

Pea pods were peeled and seeds were washed thoroughly. The juice was extracted from pea seeds using a laboratory scale juicer. A small amount of potassium metabisulfite ($K_2S_2O_5$) was added to avoid development of browning in the juice. The juice was filtered through muslin cloth. The residue left on the muslin cloth was discarded. The beaker containing filtrate was kept undisturbed overnight until a clear white layer of starch was seen at the bottom. The starch was collected and dried at a temperature of 45 °C in a hot air cabinet drier.

2.4. Amylose content

Amylose content of pea starches was measured using the method of Williams, Kuzina, and Hlynka (1970).

2.5. Scanning electron microscopy (SEM)

The micrograph was obtained by scanning electron microscopy (Joel JSM-6100, Joel LTD, Tokyo, Japan). Starch samples were suspended in ethanol to obtain a 1% suspension. A drop of starch-ethanol solution was applied onto double-backed scotch tape attached to a specimen aluminium stub and the starch was coated with gold-palladium (60:40). An accelerating potential of 10 kV was used during micrography.

2.6. Differential scanning calorimetery (DSC)

Thermal properties of starches separated from different pea cultivars were analyzed using a DSC-821 (Mettler Toledo, Switzerland) equipped with a thermal analysis data station. Starch was weighed into a 40 μl capacity aluminium pan (Mettler, ME-2733) and distilled water was added with the help of a Hamilton microsyringe to achieve a starch–water suspension containing 70% water. Samples were hermetically sealed and allowed to stand for 1 h at room temperature before heating in the DSC. The DSC analyzer was calibrated using indium and an empty aluminium pan was used as reference. Sample pans were heated at a rate of 10 °C/min from 20 to 120 °C. Onset temperature (To), peak temperature (Tp), conclusion temperature (Tc) and enthalpy of gelatinization (ΔH_{gel}) were calculated automatically. Because the peak was symmetrical, the gelatinization range was calculated on a dry starch basis. The peak height index (PHI) was calculated by the ratio ΔH_{gel}/Tp–To, as described by Krueger, Knutson, Inglett, and Walker (1987).

2.7. Turbidity

Turbidity of starches from different pea cultivars was measured as described by Perera and Hoover (1999). A 2% aqueous suspension of starch from each pea cultivar was heated in a water bath for 1 h with constant stirring and then the suspension was cooled for 1 h at 30 °C. The samples were stored for 5 days at 4 °C in a refrigerator and turbidity was determined after every 24 h by measuring absorbance at 640 nm with a Shimadzu UV-1601 spectrophotometer (Shimadzu Corporation, Kyoto, Japan).

2.8. Statistical analysis

The data reported in all the Tables are averages of quadruplicate observations. The data were subjected to analysis of variance using Minitab Statistical Software (Minitab Inc., USA).

3. Results and discussion

3.1. Seed hardness

The seed hardness test, performed on the seeds of different pea cultivars, showed that NDVP-12 cultivar

had the highest seed hardness followed by VL-7, MA-6 and Arkel cultivars. Seed hardness of seeds of different pea cultivars increased progressively during storage up to the 3rd week. However, the seeds of MA-6 and VL-7 pea cultivars showed an increase in seed hardness up to the 2nd week of storage; thereafter, a decrease was observed (Table 1). Moisture contents of the seeds from the different pea cultivars decreased during storage (Table 1). The change in seed hardness in different pea cultivars may be attributed to loss of moisture. The differences in seed hardness among pea cultivars may be attributed to differences in size and structure of starch granules. The pea cultivars with highest starch showed lowest seed hardness and vice versa.

3.2. Amylose content

The amylose contents of starches separated from different pea cultivars were in the range 26.9–61.5%. MA-6 pea starch had the lowest amylose content while Arkel pea starch had the highest amylose content. The amylose contents of MA-6, NDVP-12, VL-7 and Arkel pea starches were 26.9%, 58.5%, 61.4%, and 61.5%, respectively. These values were much higher than those reported earlier (Chavan, Shahidi, Hoover, & Perera, 1999; Czuchajowska, Otto, Paszczynska, & Baik, 1998; Ratnayake et al., 2001) for other legumes, except for MA-6 pea starch, which has a value comparable to that reported by Chavan et al. (1999).

3.3. Scanning electron microscopy (SEM)

Scanning electron microscopy depictions of starch granules from different pea cultivars differed significantly in size and shape (Fig. 1). MA-6 pea starch had larger size granules than VL-7, Arkel and NDVP-12 pea starches. The diameter of granule ranged from 18.18 to 31.81 μm in MA-6 pea starch, 3.63 to 24.54 μm inVL-7 and 2.72 to 18.18 μm in both Arkel and NDVP-12 pea starches. The values were higher for the longer diameter (31.81 μm) of MA-6 pea starch except of VL-7 (24.54 μm), Arkel and NDVP-12 (18.18 μm) pea starches which had lower values for the longer diameter. Similar longer diameter values for the field pea starches were reported by Ratnayake et al. (2001). But, for shorter diameter the values are lower for VL-7 (3.63 μm), Arkel and NDVP-12 (2.72 μm) pea starches than those reported by Ratnayake et al. (2001) for the field pea starches (10 μm). MA-6 pea starch, showed highest values for shorter diameters (18.18 μm), as well as the presence of a greater proportion of large-size granules with deeper indentations and fissures. Otto, Baik, and Czuchajowska (1997a) observed similar shaped granules in pea starch. Ratnayake et al. (2001) reported the presence of irregular granules with smooth surface and without fissures in the field pea starches. VL-7, Arkel and NDVP-12 pea starches showed the presence of granules with complicated structures and with deep fissures and grooves. Some controversy over the shape of granules of pea starch, whether compound or simple in structure, has been reported (Colonna, Gallant, & Mercier, 1980; French, 1984; Kooistra, 1962). Otto et al. (1997a) reported the structure of granules in pea starch as "broken granules". Arkel and NDVP-12 pea starch granules showed both irregular and complex structures. The deep fissures in the starch granules have been reported to be indicative of a strong bond between the starch and the protein matrix (Colonna et al., 1980). The variation in size and shape may be due to a difference in biological origin (Svegmark & Hermansson, 1993). The morphology of starch granules depends on the biochemistry of the chloroplast or amyloplast, as well as physiology of the plant (Badenhuizen, 1969).

Table 1
The effect of moisture content and compression force on seeds from different pea cultivars[a]

Variety	Storage duration (week)	Moisture content (%)	Seed hardness (N)
MA-6	1	73.06cde	7.34a
	2	64.10a	11.51bc
	3	67.60ab	21.92g
VL-7	1	74.11cde	13.93cd
	2	69.34bc	24.37g
	3	72.11cd	34.44I
Arkel	1	75.53cde	9.53ab
	2	74.48cde	15.10de
	3	74.00de	16.86ef
NDVP-12	1	77.72e	14.28de
	2	73.06cde	18.63f
	3	71.71cd	29.72h

[a] Values with similar letters in column do not differ significantly ($P < 0.05$).

3.4. Differential scanning calorimetery (DSC)

DSC parameters of starches separated from different pea cultivars are summarized in Table 2. The transition temperatures (To, Tp, Tc), range (Tc–To), enthalpy of gelatinization (ΔH_{gel}) and peak height indices (PHI) of starches from the different pea cultivars differ significantly. MA-6 pea starch showed the highest To (58.67 °C) and VL-7 pea starch showed the lowest To (50.14 °C). MA-6 and Arkel pea starches showed the highest Tc values of 69.9 °C and 69.75 °C, respectively, and VL-7 pea starch had the lowest To (51.90 °C). Arkel pea starch showed the highest Tp (64.43 °C) and VL-7 pea starch showed the lowest Tp (50.64 °C). MA-6 pea starch had the highest ΔH_{gel}, which may be due to the presence of many large-size and irregular granules. NDVP-12 pea starch showed the lowest ΔH_{gel} because it contained small size oval granules. Granule size and shape and phosphate esters have been reported to affect

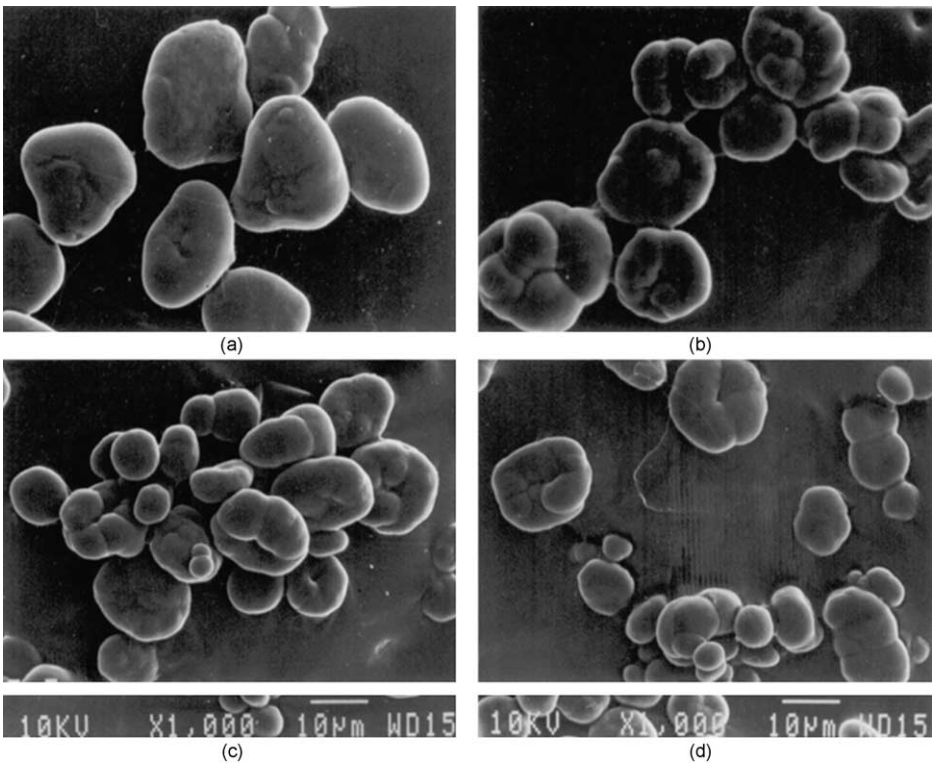

Fig. 1. Scanning electron micrographs (SEM) of starches separated from different pea cultivars (A) MA-6, (B) VL-7, (C) Arkel, (D) NDVP-12.

Table 2
Thermal properties of starches separated from different pea cultivars[a]

Variety	To	Tp	Tc	ΔHgel	PHI	R
MA-6	58.67c	63.61bc	69.90c	7.53b	1.524c	11.23b
VL-7	50.14a	50.64a	51.90a	0.83a	0.330b	1.76a
Arkel	57.90c	64.43c	69.75c	0.54a	0.082a	11.85b
NDVP-12	54.38b	63.09b	68.40b	0.11a	0.012a	14.02c

[a] Values with similar letters in column do not differ significantly ($P < 0.05$).

ΔH_{gel} values of starches (Stevens & Elton, 1971; Yuan, Thompson, & Boyer, 1993). Ratnayake et al. (2001) and Hoover and Sosulski (1991) have reported much higher To, Tp, Tc, ΔH_{gel} and range (R) for the field pea starches than observed in the present study. PHI is the ratio of ΔH_{gel} for gelatinization to the gelatinization temperature range and is a measure of uniformity in gelatinization. PHI of MA-6 pea starch was highest and that of NDVP-12 pea starch was lowest. The higher value of MA-6 pea starch may be attributed to the presence of large-size granules and the small value of the PHI of NDVP-12 pea starch may be attributed to its small-size granules. Gelatinization causes the disruption of crystalline structure in starches. Arkel and NDVP-12 pea starches showed higher R values but lower PHI. The differences in the To, R and ΔH_{gel} among pea starches may be attributed to the variation in amounts of longer chains of amylopectin. Longer chains require much higher temperatures to break completely than shorter double helices (Yamin, Lee, Pollak, & White, 1999). The ΔH_{gel} reflects the loss of double-helical rather than crystalline order (Cooke & Gidley, 1992). High transition temperature has been reported to result from a high degree of crystallinity (Barichello, Yada, Coffin, & Stanley, 1990).

3.5. Turbidity

Turbidity values of the starch from different cultivars of pea differed significantly. NDVP-12 pea starch showed highest turbidity and MA-6 pea starch showed lowest turbidity. The turbidity values observed for pea starch pastes, in the present study, were higher than those observed earlier for potato starches under similar experimental conditions (Kaur, Singh, & Sodhi, 2002). These differences may be attributed to differences in phosphate monoester contents. Phosphate monoesters have been reported to affect the starch paste clarity (Craig, Maningat, Seib, & Hoseney, 1989). The value of turbidity for MA-6, VL-7 and Arkel pea starch pastes increased progressively up to the 3rd day of storage and, thereafter non-significant changes were observed. NDVP-12 pea starch showed an increase only up to the 2nd day of storage and then the turbidity was not affected significantly. MA-6 pea starch showed the lowest turbidity, which may be attributed to the presence of

Table 3
Effect of storage duration on turbidity values of starch pastes from different pea cultivars[a]

Variety	1st Day	2nd Day	3rd Day	4th Day	5th Day
MA-6	1.927a	2.737a	2.767a	2.767a	2.767a
VL-7	2.872b	3.010b	3.068b	3.135b	3.135b
Arkel	2.960b	3.010b	3.068b	3.135b	3.135b
NDVP-12	3.068b	3.135b	3.135b	3.135b	3.135b

[a] Values with similar letters in column do not differ significantly ($P < 0.05$).

large size granules, while NDVP-12 pea starch, with small granules, showed higher turbidity. The turbidities of Arkel and VL-7 pea starches did not differ significantly. Factors such as granule swelling, granule remnants, leached amylose and amylopectin, amylose and amylopectin chain length, intra-or inter-molecular bonding and lipid cross-linking substitution have been reported to be responsible for turbidity development in starches during storage (Jacobson, Obanni, & BeMiller, 1997) (Table 3).

4. Conclusion

The Indian pea cultivars (MA-6, VL-7, NDVP-12, Arkel) differ significantly in seed hardness and starch properties. The pea cultivars with greater seed hardness had smaller starch granules than those with lower seed hardness. Starches isolated from the different cultivars differed significantly in turbidity, amylose content, morphological and thermal properties. MA-6 pea starch, having large size granules, had lowest amylose content and turbidity and highest ΔH_{gel}, PHI and R.

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