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# Microstructure, physicochemical properties and *in vitro* digestibility of starches from different Indian lentil (*Lens culinaris*) cultivars

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#### ABSTRACT

Starches isolated from four *Lens culinaris* cultivars were evaluated for their physicochemical, structural, thermal, pasting and *in vitro* digestibility characteristics. Amylose content of the *L. culinaris* starches from different cultivars varied from 30.6% to 33.9% and the degrees of crystallinity were 27.5–33.1%, with X-ray diffraction pattern of the C-type. *Lens culinaris* starch granules were oval to spherical shaped with a smooth surface and mean particle diameter of 15.9–17.4  $\mu$ m. The transition temperatures and enthalpy of gelatinization ( $\Delta H_{\rm gel}$ ) ranged between 60.2–61.3, 67.6–68.7, 74.5–75.6 °C and 8.36–8.52 J/g, respectively. The amounts of rapidly digesting, slowly digesting and resistant starch contents of *L. culinaris* starches ranged from 56.0 to 65.5%, 5.1% to 9.2%, and 29.4% to 34.8%, respectively. Digestibility and  $\Delta H_{\rm gel}$  showed a statistically significant correlation with amylose content, relative crystallinity and particle diameter. All the four starches exhibited nearly identical pasting temperatures but differed significantly (P<.05) with respect to peak, breakdown, final and setback viscosity.

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#### 1. Introduction

Legumes play an important role in the diet of most of the people of the world and are second only to the cereals as a source of human and animal food (Singh, Kaur, Sandhu, & Guraya 2004a). They are excellent source of carbohydrates (50-60%) and an inexpensive source of proteins (20-24%). Commonly consumed legumes in Indian diets are lentil, green gram, black gram, bengal gram, kidney bean and soya. The lentil (Lens culinaris) is a bushy annual plant of the legume family, grown for its lens-shaped seeds. Lentils are relatively tolerant to drought and are grown throughout the world. Lentil growing is concentrated mostly in the semi-arid regions in the Indian subcontinent, the Mediterranean regions and the dry areas of the Middle East. The total world production of lentils was 3.874 million metric tonnes (FAO, 2007), with India contributing about 36% of the total production. A large percentage of Indians are vegetarian and lentils have long been part of the indigenous diet as a common source of protein.

A variety of lentils exists with colours that range from yellow to red-orange to green, brown and black. Lentils are divided into two main types based on difference between the seed coat and cotyledon colour. Green lentils (Macrosperma) have a green to brown seed coat with yellow cotyledons. Red lentils (Microsperma) have a pale grey to dark seed coat with red cotyledons. Like most

legumes, lentil seeds contain about two-thirds carbohydrates, 24–30% protein and are also a good source of B-complex vitamins, such as folate, thiamine, niacin and riboflavin, with a good balance of minerals (Jood, Bishnoi, & Sharma, 1998; Longnecker, Kelly, & Huang, 2002). Lentil seeds provide an excellent source of dietary fiber and complex carbohydrates (Sotomayor et al. 1999).

Starch is the major storage polysaccharide of higher plants and is deposited in partially crystalline granules varying in morphology and molecular structure between and within plant species (Blazek & Copeland, 2008). Starch is of considerable commercial importance because of its numerous desirable functional properties, especially related to its ability to modify texture of a product. Sotomayor et al. (1999) and Rosin, Lajolo, and Menezes (2002) reported that raw lentil has about 51% and 59% (dry matter) starch concentration, respectively. An important property of starch in relation to its functionality is its ability to absorb water, resulting in gelatinization and loss of granular organization (Blazek & Copeland, 2008). Differences in starch physicochemical characteristics have significant impact on their functional and rheological behaviour, affecting their suitability for specific uses.

Starch is not completely digested and absorbed in the small intestine, as was previously thought before the early 1980s. Englyst, Kingman, and Cummings (1992) first recognized the presence of starch fraction resistant to enzymic hydrolysis during their research on measurement of non-starch polysaccharides. Most starch products contain a portion that digests rapidly (rapidly digesting starch, RDS), a portion that digests slowly (slowly

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digesting starch, SDS) and, a fraction that is resistant to digestion (resistant starch, RS) (Englyst et al. 1992). RS has potential physiological benefits similar to dietary fiber and unique functional properties. Legumes have been shown to contain significant amounts of RS which is especially beneficial for reducing the risk of several diseases. The digestibility of starch in foods varies widely and is greatly influenced by plant type, processing conditions, physicochemical characteristics of the starch and plant microstructure and composition (Ring, Gee, Whittam, Orford, & Johnson 1988).

Previous studies on lentil starches have been focused primarily on its physicochemical and functional properties (Hoover & Manuel, 1995; Hoover & Ratnayake, 2002; Shahen, Roushdi, & Hassan, 1977; Yoshimi & Toshiko, 2006). Investigations have also been carried out to see the impact of annealing and heat-moisture treatment on digestibility behaviour of native and gelatinized lentil starches (Chung, Liu, & Hoover, 2009), Chung et al. (2008) compared the molecular and digestibility characteristics of pea, lentil and chickpea cultivars of Canada. However, there is dearth of information on relationship between starch digestibility, and it's structural and physical properties among different lentil cultivars. Therefore, the present study was focused on determination of functional, structural and in vitro digestibility characteristics of starches separated from the four L. culinaris cultivars grown in India and were related to rheological characteristics like swelling, gelatinization and pasting. Functionality and rheological characterization can be used for selection of appropriate lentil cultivars to see their suitability in different food processes.

#### 2. Materials and methods

#### 2.1. Materials

Representative samples of four improved red lentil (*L. culinaris*) cultivars viz. (LL-912, LL-699, LL-56 & LL-147) were obtained from Punjab Agricultural University, Ludhiana, India. The samples were estimated for their moisture, ash, fat, and protein content by employing the standard methods of analysis (AOAC, 1984). Starches were isolated from *L. culinaris* cultivars following the method as described by Singh et al. (2004a).

# 2.2. Physicochemical properties

#### 2.2.1. Amylose content

Amylose content of isolated *L. culinaris* starches was determined by following the method of Williams, Kuzina, and Hlynka (1970).

### 2.2.2. Swelling power (g/g) and solubility (%)

Swelling power and solubility were determined in triplicate using method of Leach, McCowen, and Schoch (1959).

#### 2.3. Size distribution

Size distribution of *L. culinaris* starches was measured using a Laser light scattering particle size analyzer (1064 LD, CILAS, France) following the method described by Singh et al. (2004a).

#### 2.4. Morphological properties

Scanning electron micrographs were taken by a Jeol JSM-6100 Scanning Electron Microscope (Jeol Ltd., Tokyo, Japan). Starch samples were suspended in ethanol to obtain a 1% suspension. One drop of the starch–ethanol suspension was applied on an aluminium stub using double-sided adhesive tape and the starch was coated with gold–palladium (60:40). An accelerating potential of 15 kV was used during micrography.

#### 2.5. X-ray diffraction analysis

X-ray diffraction analysis was conducted using an X-ray diffractometer (Philips, X'pert MPD high resolution XRD, Almelo, Netherlands) operated at 40 kV and 40 mA. Diffractograms were obtained from  $4^{\circ}$  ( $2\theta$ ) to  $30^{\circ}$  ( $2\theta$ ) at a scanning speed of  $4^{\circ}$ /min. The degree of relative crystallinity was determined quantitatively following the method described by Nara and Komiya (1983) using peak-fitting software (Origin – Version 6.0, Microcal Inc., Northampton, MA, USA).

#### 2.6. Thermal properties

Thermal properties of isolated *L. culinaris* starches were analyzed using a Differential Scanning Calorimeter (Seiko Instrument, DSC 6100, Chiba, Japan) following the method described by Singh, Sandhu, and Kaur (2004b). After conducting thermal analysis, samples were stored in the refrigerator at 4  $^{\circ}$ C for 7 days for retrogradation studies. Retrogradation was measured by reheating the sample pans containing the starches at the rate of 10  $^{\circ}$ C/min from 25 to 100  $^{\circ}$ C.

#### 2.7. Pasting properties

The pasting properties of *L. culinaris* starches were evaluated using a Rapid Visco Analyzer (RVA-3D, Newport Scientific, Warriewood, Australia) following the method described by Singh et al. (2004b). Viscosity profiles of starches were recorded using starch suspensions (8%, w/w; 28 g total weight).

#### 2.8. In vitro starch digestibility

In vitro starch digestibility was analyzed following the method described by Englyst et al. (1992) with slight modifications (Sandhu & Lim, 2008a). Amyloglucosidase (No. 9913, Sigma–Aldrich) (1 ml) was added to deionized water (2 ml). Porcine pancreatic alpha-amylase (No. 7545, Sigma–Aldrich, St. Louis, MO) (3.89 g) was dispersed in water (25.7 ml) and centrifuged for 10 min at 2500g, and 18.7 ml of supernatant was collected. This supernatant was mixed with 1 ml of diluted amyloglucosidase for making the enzyme solution. The solution was freshly prepared for the digestion analysis.

Aliquots of guar gum (10 ml, 5 g/l) and sodium acetate (5 ml, 0.5 M) were added to the starch samples (0.5 g, db) in a test tube. Seven glass balls (10 mm diameter) and 5 ml of enzyme solution were then added to each tube, following the incubation in a water bath (37 °C) with agitation (170 rpm). Aliquots (0.5 ml) were taken at intervals and mixed with 4 ml of 80% ethanol, and the glucose contents in the mixture were measured using glucose oxidase and peroxidase assay kits (No. GAGO-20, Sigma–Aldrich). The total starch content was measured according to Englyst et al. (1992). The starch classification based on its digestibility was: RDS as the starch that was hydrolyzed within 20 min of incubation, RS as the starch not hydrolyzed with 120 min, and SDS as the starch digested during the period between 20 and 120 min.

# 2.8.1. Estimated glycemic index (GI)

Using the hydrolysis curve (0–180 min), the hydrolysis index (HI) was calculated as the percentage of total glucose released from the sample compared to that released from white bread (Goñi, Garcia-Alonso, & Saura-Calixto, 1997; Granfeldt, Björck, Drews, & Towar 1992). The glycemic indices of the samples were estimated according to the equation described by Goñi et al. (1997): GI = 39.71 + 0.549 HI.

**Table 1**Physicochemical properties, mean particle diameter and relative crystallinity of starches from different *Lens culinaris* cultivars. xy

Parameters	LL-912	LL-699	LL-56	LL-147
Moisture (%)	$9.2 \pm 0.2^{ab}$	$8.9 \pm 0.3^{a}$	9.1 ± 0.1 <sup>ab</sup>	9.4 ± 0.2 <sup>b</sup>
Ash (%)	$0.19 \pm 0.02^{a}$	$0.26 \pm 0.04^{\circ}$	$0.22 \pm 0.05^{b}$	$0.25 \pm 0.04$ <sup>bc</sup>
Fat (%)	$0.10 \pm 0.01^{a}$	$0.11 \pm 0.01^{ab}$	$0.09 \pm 0.01^{a}$	$0.14 \pm 0.02^{b}$
Protein (%)	$0.69 \pm 0.09^{a}$	$0.74 \pm 0.1^{b}$	$0.72 \pm 0.2^{ab}$	$0.81 \pm 0.08^{c}$
Amylose content (%)	$31.2 \pm 0.6^{ab}$	$32.5 \pm 0.7^{b}$	$30.6 \pm 0.5^{a}$	$33.9 \pm 0.5^{\circ}$
Swelling power (g/g)	13.7 ± 0.4 <sup>b</sup>	$11.9 \pm 0.3^{a}$	$13.6 \pm 0.5^{b}$	$12.8 \pm 0.4^{ab}$
Solubility (%)	$14.4 \pm 0.4^{b}$	$13.5 \pm 0.3^{a}$	13.1 ± 0.2 <sup>a</sup>	$13.8 \pm 0.3^{ab}$
Mean particle diameter (µm)	16.5 <sup>b</sup>	16.1 <sup>ab</sup>	17.4 <sup>c</sup>	15.9 <sup>a</sup>
Relative crystallinity (%)	30.5 ± 0.3 <sup>a</sup>	28.1 ± 0.2 <sup>ab</sup>	$33.1 \pm 0.2^{\circ}$	$27.5 \pm 0.4^{a}$

 $<sup>^{\</sup>rm x}$  Means followed by same superscript within a row do not differ significantly (P < .05).

#### 2.9. Statistical analysis

The data reported in all the tables were average of triplicate observations. The data were subjected to one-way analysis of variance (ANOVA) using Minitab Statistical Software version 13 (Minitab Inc., USA). Pearson correlation coefficients (r) for the relationships between various starch properties were calculated.

#### 3. Results and discussion

#### 3.1. Physicochemical properties

The chemical composition of starches separated from the different L. culinaris cultivars are presented in Table 1. The isolated L. culinaris starches were characterized by low ash, protein and fat content of 0.19-0.26%, 0.69-0.81% and 0.09-0.14%, respectively. The low ash, fat and protein contents of the extracted starches reflect the purity of the starches extracted from the L. culinaris cultivars. The amylose content of the *L. culinaris* cultivar starches was 30.6–33.9% (Table 1), agreeing with the previously reported values of 33.0-34.5% (Hoover & Manuel, 1995) and 30.1-34.4% (Yoshimi & Toshiko, 2006) in lentil starches. Lens culinaris cv. LL-147 starch had the highest amylose content while cv. LL-56 starch had the lowest value for the same. Amylose content has a significant effect on functional and physicochemical properties, including pasting, gelatinization, retrogradation and swelling behaviour of starch (Blazek & Copeland, 2008; Syegmark et al. 2002; Whistler & BeMiller, 1996).

The ability of the starches from the different *L. culinaris* cultivars to swell in excess of water and their solubility is presented in Table

1. Swelling power (SP) and solubility of the four *L. culinaris* starches ranged from 11.9 to 13.7 g/g and 13.1% to 14.4%, respectively. *Lens culinaris* cv. LL-912 starch showed higher SP and solubility than the other lentil starches. SP and solubility provide an evidence of the magnitude of interaction between starch chains within the amorphous and crystalline domains. The extent of this interaction has been reported to be influenced by the amylose/amylopectin ratio (Blazek & Copeland, 2008), strongly bonded micellar network (Gujska, Reinhard, & Khan, 1994) and amylopectin molecular structure (Tester, Morrison, & Schuiman, 1993). A negative correlation, though not statistically significant, of SP with amylose content was observed.

#### 3.2. Crystallinity

X-ray diffraction was used to study the presence and characteristics of the crystalline structure of the starch granules. In the diffraction spectra of the four starches from L culinaris cultivars, there were three strong diffraction peaks at  $15^{\circ}$ ,  $17.2^{\circ}$  and  $23.2^{\circ}$  ( $2\theta$ ). These showed a C-type X-ray diffraction pattern for lentil starches. A C-type X-ray pattern for different legume starches has been previously reported (Hoover & Ratnayake, 2002). The degree of crystallinity of the four L culinaris starches ranged between  $27.5^{\circ}$  and  $33.1^{\circ}$  (Table 1). Comparable relative crystallinity values ranging between  $27.1^{\circ}$  and  $33.5^{\circ}$  (Zhou, Hoover, & Liu, 2004) for C-type legume starches and  $26.2-28.3^{\circ}$  for lentil starches (Chung et al., 2008) have been reported earlier. Relative crystallinity was negatively correlated to amylose content at a significance level of P < .01 (Table 2). Since the side chains of amylopectin form the crystalline structure, it is expected that the crystallinity will be

 Table 2

 Pearson correlation coefficient between various properties of starches from different Lens culinaris cultivars.

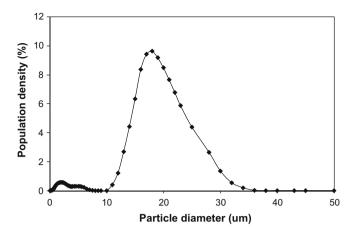
	Aml	SP	Dia	Crys	To	$\Delta H_{ m gel}$	PT	PV	BV	FV	RS	RDS	SDS
SP	-0.633												
Dia	$-0.898^{*}$	0.553											
Crys	$-0.926^{*}$	0.749	0.965**										
$T_{\rm o}$	-0.420	-0.437	0.401	0.202									
$\Delta H_{\rm gel}$	0.901*	-0.322	-0.944**	$-0.859^{*}$	-0.672								
PT	0.127	$-0.846^{*}$	-0.126	-0.348	$0.844^{*}$	-0.181							
PV	-0.047	0.843*	0.014	0.248	$-0.887^{*}$	0.283	-0.993**						
BV	-0.184	0.877	0.134	0.369	-0.815	0.157	-0.993**	0.990**					
FV	0.915*	-0.266	-0.825	-0.759	-0.751	0.950**	-0.285	0.361	0.229				
RS	-0.970**	0.796	0.895*	0.968**	0.195	-0.818	-0.360	0.279	0.408	-0.791			
RDS	0.954**	-0.805	$-0.917^{*}$	-0.985**	-0.167	0.820	0.388	-0.301	-0.427	0.766			
SDS	0.976**	-0.785	$-0.874^{*}$	-0.951**	-0.214	0.812	0.338	-0.261	-0.391	0.806	-0.998**	0.989**	
GI	-0.232	-0.538	0.048	-0.098	0.895*	-0.373	0.875*	$-0.865^{*}$	-0.816	-0.573	-0.011	0.069	-0.033

Aml, amylose content; SP, swelling power; Dia, particle diameter; Crys, relative crystallinity;  $T_0$ , onset gelatinization temperature;  $\Delta H_{\rm gel}$ , enthalpy of gelatinization; PT, pasting temperature; PV, peak viscosity; BV, breakdown viscosity; FV, final viscosity; RDS, readily digestible starch; SDS, slowly digestible starch; RS, resistant starch; GI, glycemic index.

y Mean (±standard deviation) of triplicate analysis.

<sup>\*</sup> P < .05.

<sup>\*\*</sup> *P* < .01.



**Fig. 1.** Particle size distribution of starches separated from the four *Lens cultivars*.

inversely related to amylose content (Sandhu & Lim, 2008b). The crystalline order in starch granules is often the basic underlying factor influencing its functional properties.

#### 3.3. Morphological properties and size distribution

The starches separated from the four *L. culinaris* cultivars differed significantly in their granule size. Fig. 1 shows the granule size distribution of the four *L. culinaris* cultivar starches. Particle

diameter of the majority of the starch granules ranged between 11 and 36 µm with a few granules having diameter in the range of 1-7.5 μm. The average particle diameter of the *L. culinaris* starches ranged between 15.9 and 17.4 µm, the largest for L. culinaris cv. LL-56 starch and the lowest for cv. LL-147 starch (Table 1). A negative correlation of particle diameter with amylose content (r = -.898) and a positive correlation with relative crystallinity (r = .965) at a significance level of P < .01 was observed. Similar observation on influence of granule size on amylose content of corn starches has been observed earlier (Sandhu, Singh, & Kaur, 2004). Starch granule size may affect its physicochemical properties, such as gelatinization and pasting, enzyme susceptibility, crystallinity, and solubility (Lindeboom, Chang, & Tyler, 2004). The scanning electron micrographs (SEM) of starches separated from the four L. culinaris cultivars showed the presence of large oval to spherical shape granules (Fig. 2). The surface of the granules appeared to be smooth with no evidence of any fissures. Lentil starch granules have been reported to have round to elliptical shaped granules, with characteristic dimensions in the range of 2.5-25 µm (Hoover & Manuel, 1995) and 16-19 µm (Yoshimi & Toshiko, 2006).

#### 3.4. Thermal properties

DSC was used to study starch gelatinization which involved disruption of the native structure of the starch granule. The results of DSC analysis of the starches separated from the four L. culinaris cultivars are presented in Table 3. Significant differences were observed in transition temperatures ( $T_0$ ,  $T_p$  and  $T_c$ ), enthalpy of

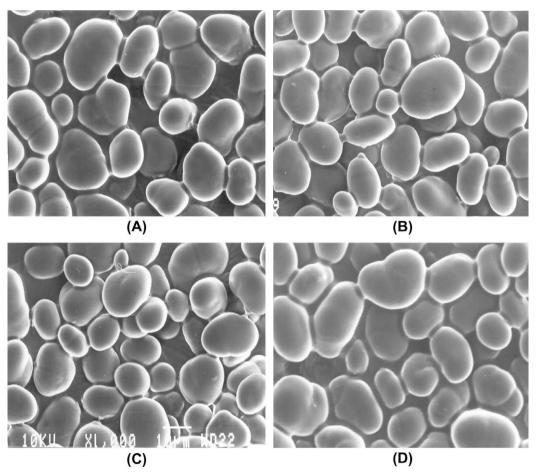


Fig. 2. Scanning electron micrographs of starches separated from the four Lens culinaris cultivars: (A) LL-912, (B) LL-699, (C) LL-56, and (D) LL-147.

**Table 3**Thermal properties of starches from different *Lens culinaris* cultivars.\*xy

Parameter	LL-912	LL-699	LL-56	LL-147
$T_{o}$ (°C) $T_{p}$ (°C) $T_{c}$ (°C) $\Delta H_{gel}$ (J/g)	$60.6 \pm 0.3^{ab}$ $68.3 \pm 0.2^{ab}$ $75.4 \pm 0.3^{b}$ $8.44 \pm 0.4^{ab}$	$61.3 \pm 0.4^{b}$ $68.7 \pm 0.5^{b}$ $75.6 \pm 0.4^{b}$ $8.43 \pm 0.2^{ab}$	$60.9 \pm 0.3^{ab}$ $67.9 \pm 0.3^{a}$ $75.1 \pm 0.2^{ab}$ $8.36 \pm 0.2^{a}$	$60.2 \pm 0.2^{a}$ $67.6 \pm 0.3^{a}$ $74.5 \pm 0.5^{a}$ $8.52 \pm 0.3^{b}$
R ΔH <sub>ret</sub> (J/g) %R	$14.8 \pm 0.4^{a}$ $5.93 \pm 0.2^{a}$ $70.26 \pm 0.6^{a}$	$14.3 \pm 0.5^{a}$ $5.99 \pm 0.2^{b}$ $71.06 \pm 0.5^{b}$	$14.2 \pm 0.4^{a}$ $6.04 \pm 0.3^{c}$ $72.25 \pm 0.5^{c}$	$14.3 \pm 0.3^{a}$ $5.98 \pm 0.2^{b}$ $70.18 \pm 0.5^{a}$

 $T_{\rm o}$ , onset gelatinization temperature;  $T_{\rm p}$ , peak gelatinization temperature;  $T_{\rm c}$ , conclusion gelatinization temperature; R, gelatinization range  $(T_{\rm c}-T_{\rm o})$ ;  $\Delta H_{\rm gel}$ , enthalpy of gelatinization (dwb, based on starch weight);  $\Delta H_{\rm ret}$ , enthalpy of retrogradation; R, ratio of enthalpy of retrogradation to enthalpy of gelatinization.

gelatinization ( $\Delta H_{gel}$ ) and gelatinization temperature range (R) of starches from L. culinaris cultivars. The transition temperatures of L. culinaris starches ranged from 60.2 to 61.3, 67.6 to 68.7, 74.5 to 75.6 °C, respectively, in agreement with the previous reports in chickpea (Singh et al. 2004b), black gram (Singh et al. 2004a) and different legume starches (Sandhu & Lim, 2008a). The differences in gelatinization temperature may be attributed to the differences in amylose content, size, form and distribution of starch granules, and to the internal arrangement of starch fractions within the granule. Lens culinaris cv. LL-699 showed the highest  $T_0$ ,  $T_p$ ,  $T_c$ whereas the lowest was observed for cv. LL-147 starch. To was positively correlated to  $T_p$  (r = .815) and  $T_c$  (r = .836).  $\Delta H_{\rm gel}$  of starches from different L. culinaris cultivars varied from 8.36 to 8.52 J/g. Lens culinaris cv. LL-147 starch showed the highest  $\Delta H_{\rm gel}$ , which may be due to its lowest degree of crystallinity.  $\Delta H_{\rm gel}$  exhibited a statistically significant negative correlation with particle diameter (r = -.944) and relative crystallinity (r = -.859) (Table 2). The gelatinization enthalpies have been related to characteristics of the starch granule such as degree of crystallinity (Krueger, Knutson, Inglett, & Walker 1987) and granule size (Bogracheva, Meares, & Hedley, 2006). The crystallinity of starch decreases with gelatinization and a completely gelatinized starch has an amorphous structure. The gelatinization temperature range of the L. culinaris starches varied between 14.2 and 14.8 °C. The gelatinization properties of starch are related to a variety of factors including the size, proportion and kind of crystalline organization, and ultra structure of the starch granule (Lindeboom et al., 2004).

The retrogradation properties of the *L. culinaris* starches were studied after storage of gelatinized starches at 4 °C for 7 days. Transition temperatures and retrogradation enthalpy ( $\Delta H_{\rm ret}$ ) at the end of the storage period dropped significantly, compared to transition temperatures and  $\Delta H_{\rm gel}$  during gelatinization (Table 3). Starch retrogradation enthalpies are usually smaller (60–80%) in comparison to gelatinization enthalpies because of weaker starch crystallinity in retrograded starch (Sasaki, Yasui, & Matsuki, 2000).  $\Delta H_{\rm ret}$  for the four *L. culinaris* cultivar starches ranged between 5.93 and 6.04 J/g. In retrograded starches, the enthalpy provides a quantita-

tive measure of the energy transformation that occurs during melting of recrystallized amylopectin as well as precise measurements of the transition temperatures of the endothermic event (Karim, Norziah, & Seow, 2000). The difference in  $\Delta H_{\rm ret}$  among the various L. culinaris starches suggested difference in their tendency towards retrogradation. Starch from *L. culinaris* cv. LL-56 had a higher  $\Delta H_{\rm ret}$  and % retrogradation (6.04 J/g and 72.25%, respectively) in comparison to other lentil starches. Greater retrogradation tendency of starch has traditionally been linked to a greater amylose content (Whistler & BeMiller, 1996), but amylopectin and intermediate materials also play a significant role in starch retrogradation during refrigerated storage (Yamin, Lee, Pollak, & White 1999).

#### 3.5. Pasting properties

Rapid Visco Analyzer (RVA) is an effective instrument for measuring the viscous properties of cooked starch and for relating functionality to structural properties (Blazek & Copeland, 2008). Significant differences (P < .05) in the pasting properties between the L. culinaris cultivar starches were observed (Table 4). All the four L. culinaris starches exhibted nearly identical pasting temperatures (PT) of 74.3-74.4 °C, except for cv. LL-699 starch which had PT of 75.1 °C. Lentil starches have been reported to have PT of 75-83 °C (Hoover & Manuel, 1995). However, the L. culinaris starches differed significantly with respect to peak (2502-2787 cP), breakdown (416-757 cP), final (3385-3632 cP) and setback (1353–1562 cP) viscosity. Lens culinaris cv. LL-912 starch exhibited the highest peak, breakdown and the lowest trough viscosities, while the cv. LL-147 starch showed the highest values for final and setback viscosities. Changes in viscosity during cooking period give indications of the stability and the changes occurring during cooling show the consistency of the product when consumed. The low peak, breakdown and setback viscosity of L. culinaris cv. LL-699 starch corroborates well with its low swelling power in water. It has been postulated that starches that are capable of swelling to a higher degree are also less resistant to breakdown on cooking. Pasting temperature showed significant negative correlation with SP (r = -.846, P < .05), peak (r = -.993, P < .01) and breakdown (r = -.993, P < .01) viscosity. A significant negative correlation of relative crystallinity with trough viscosity (r = -.844. P < .05) was observed. The correlation coefficients of SP with peak. and breakdown viscosity were 0.843 and 0.877, respectively (Table 2). A positive correlation of  $\Delta H_{gel}$  with final and setback viscosities (P < .01) was observed. The starch pasting properties have been shown to be influenced by granule swelling, friction between swollen granules, amylose leaching, starch crystallinity and chain length of the starch components (Rasper, 1982).

## 3.6. Digestibility studies

The rapidly digesting starch (RDS), slowly digesting starch (SDS) and resistant starch (RS) contents of starches from the four *L. culinaris* cultivars are shown in Table 5. RS accounted for 56.0–65.5%, whereas RDS and SDS represented 5.1–9.2% and 29.4–34.8%,

**Table 4**Pasting properties of starches from different *Lens culinaris* cultivars.\*\*xy

Parameter	LL-912	LL-699	LL-56	LL-147
Pasting temperature (°C) Peak viscosity (cP) Trough viscosity (cP) Breakdown viscosity (cP) Final viscosity (cP) Setback viscosity (cP)	$74.3 \pm 0.3^{a}$ $2787 \pm 12^{c}$ $2030 \pm 14^{a}$ $757 \pm 13^{d}$ $3444 \pm 17^{b}$ $1414 \pm 13^{b}$	$75.1 \pm 0.3^{b}$ $2502 \pm 17^{a}$ $2086 \pm 8^{b}$ $416 \pm 6^{a}$ $3443 \pm 21^{b}$ $1357 \pm 12^{a}$	$74.4 \pm 0.2^{a}$ $2717 \pm 19^{b}$ $2032 \pm 12^{a}$ $685 \pm 12^{b}$ $3385 \pm 11^{a}$ $1353 \pm 11^{a}$	74.3 ± 0.3 <sup>a</sup> 2786 ± 9 <sup>c</sup> 2070 ± 11 <sup>b</sup> 716 ± 9 <sup>c</sup> 3632 ± 16 <sup>c</sup> 1562 ± 11 <sup>c</sup>

 $<sup>^{\</sup>rm x}$  Means followed by same superscript within a row do not differ significantly (P < .05).

<sup>&</sup>lt;sup>x</sup> Means followed by same superscript within a row do not differ significantly (P < .05).

y Mean (±standard deviation) of triplicate analysis.

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**Table 5**Digestibility properties of starches from different *Lens culinaris* cultivars.\*xy

Parameter	LL-912	LL-699	LL-56	LL-147
RS (%)	63.5 ± 1.4 <sup>b</sup>	57.7 ± 1.3 <sup>ab</sup>	65.5 ± 0.9°	$56.0 \pm 0.7^{a}$
RDS (%)	$6.3 \pm 0.2^{b}$	$8.7 \pm 0.6^{bc}$	$5.1 \pm 0.3^{a}$	$9.2 \pm 0.5^{\circ}$
SDS (%)	$30.2 \pm 0.8^{ab}$	$33.6 \pm 0.7^{b}$	$29.4 \pm 0.5^{a}$	$34.8 \pm 0.4^{\circ}$
HI	$12.2 \pm 0.5^{ab}$	$13.0 \pm 0.3^{b}$	$11.9 \pm 0.4^{a}$	$11.4 \pm 0.6^{a}$
GI <sup>z</sup>	$46.40 \pm 0.2^{ab}$	$46.85 \pm 0.6^{b}$	$46.24 \pm 0.6^{ab}$	$45.96 \pm 0.5^{a}$

RDS, readily digestible starch; SDS, slowly digestible starch; RS, resistant starch; HI, hydrolysis index; GI, glycemic index.

- $^{\times}$  Means followed by same superscript within a row do not differ significantly (P < .05).
  - y Mean (±standard deviation) of triplicate analysis.
- $^{z}$  GI was calculated using the equation proposed by Goñi et al. (1997): GI = 39.71 + 0.549 HI.

respectively of the total starch content in L. culinaris cultivars. High RS content of lentil starches offers its potential as a food ingredient as RS possesses physiological benefits similar to dietary fiber. RDS is rapidly and completely digested in the small intestine and is associated with more rapid elevation of postprandial plasma glucose whereas SDS is more slowly digested in the small intestine and is generally the most desirable form of dietary starch (Jenkins et al. 1981). Starch from L. culinaris cv. LL-147 showed significantly higher RDS, SDS and lower RS in comparison to other L. culinaris starches. In the present study, all the four lentil starches showed higher SDS in comparison to their RDS content which is beneficial in the management of certain diseases such as diabetes, as a high level of SDS does not produce hyperglycemia. Differences in the digestibility of native starches among species have been attributed to the interplay of many factors, such as starch source (Ring et al. 1988), granule size (Lindeboom et al. 2004), amylose/amylopectin ratio (Hoover & Sosulski, 1991), retrogradation of amylose and degree of crystallinity (Chung, Lim, & Lim, 2006). Digestibility was negatively correlated to starch granule diameter at a significance level of P < .05. Size of starch granules may affect digestibility, as the relationship between surface area and starch volume, and thus contact between substrate and enzyme, decreases as the size of granule increases (Svihus, Uhlen, & Harstad, 2005). Both RDS and SDS showed a negative correlation with relative crystallinity and a positive with amylose content (P < .01) (Table 2).

#### 3.6.1. Hydrolysis index (HI) and estimated glycemic index (GI)

Hydrolysis index (HI) expresses the digestibility of the starch in foods in relation to the digestibility of starch in a reference material, namely white bread (Sandhu & Lim, 2008b). GI is greatly influenced by the starch digestibility in the food system. The HI and GI of the four *L. culinaris* cultivar starches differed significantly varying between 11.4–13.0% and 45.96–46.85%, respectively. Starch from *L. culinaris* cv. LL-147 showed significantly lower HI and GI whereas cv. LL-699 starch exhibited the highest values for the same. GI is related to nutritional quality of food and a product with a low GI is preferable not only in individuals with diabetes, but also in healthy individuals (Björck & Asp, 1994).

#### 4. Conclusion

Significant differences in morphological and physicochemical properties were observed among the four L. culinaris cultivar starches. These differences influence the observed variations in pasting properties, gelatinization parameters, and digestibility characteristics. Lens culinaris cv. LL-147 differed significantly from starches of other cultivars in exhibiting the highest amylose content,  $\Delta H_{\rm gel}$  and lowest transition temperatures, mean particle diameter and relative crystallinity. Correlation analysis data indicated a significant interdependence of physicochemical and struc-

tural properties with thermal, and digestibility properties of L culinaris starches. The digestibility and  $\Delta H_{\rm gel}$  of starch showed a significant correlation to granule diameter, relative crystallinity and amylose content. The results revealed that L culinaris starches and their fractions varied significantly in their digestibility behaviour. The low RDS, GI and the high RS and SDS content of lentil starches makes them of public health value for its use in sustained glucose release applications, particularly for diabetes management. Structure property relationships have been established for native lentil starches. It is desirable that further studies be conducted on utilization of lentil starches in common food products like starch noodles (to partially or totally substitute mung bean starch) and snack foods and their structural modification by chemical processes to make lentil starch more useful for the food processors.

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