Physicochemical Properties of Crude and Purified Locust Bean Gums Extracted from Iranian Carob Seeds

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ABSTRACT

In this research, seeds of Iranian carob trees grown wild in Kazeroon city (Fars Province) were used to extract Iranian locust bean gum as a seed galactomannan. Chemical composition, solubility, and rheological properties of crude and purified gums were investigated and compared with a commercial (Sigma) locust bean gum (LBG) sample. Chemical composition of the purified Iranian LBG was similar to the commercial LBG, however, the protein content of Iranian crude LBG was higher. Solubility data showed that the Iranian crude LBG was partially cold-water soluble (~ 55% at 25°C) and needed heating to reach a higher solubility value (~ 71% at 80 °C). Rheological properties of the solutions of the three LBG samples were investigated as a function of concentration and temperature (5-60 °C) at shear rate range of 0.5-200 s^{-1}. All three samples showed shear thinning behavior and Power law model was used to fit the experimental data. The Iranian LBG (crude and pure) was able to thicken aqueous solutions; however, its viscosity level was lower than that of the commercial LBG. Study of rheological properties of the three LBG samples at different pH levels (2.5-10) showed that they were significantly affected by pH. The intrinsic viscosities of the Iranian crude and purified LBG and commercial LBG were 3.95, 9.05, and 9.48 dL g^{-1}, respectively.

Keywords: Consistency index, Carob seeds, Iranian LBG, Intrinsic viscosity, Rheology.

INTRODUCTION

Galactomannans are hydrophilic biopolymers composed of a linear mannose backbone with varying amount of galactose substituents linked to the main backbone by glycosidic bonds (Wielinga and Maehall, 2000). Galactomannans that are extracted from plant seeds are in seed gums group and they are biodegradable and biocompatible polymers (Pazur *et al*., 1986).

One of the most important seed galactomannans in food industry is locust bean gum (LBG) (Wielinga and Maehall, 2000). Carob tree is known as the first source of seed galactomannans. This evergreen tree belongs to *Caesalpinaceae* subfamily from *Leguminaceae* family and grows in semi-arid or subtropical zones (Yousif and Alghzawi, 2000; Battle and Tous, 1997). Locust bean gum is the milled and purified endosperm of seeds of carob tree that is mainly used in food and non-food products as thickening and stabilizing agents. Degree of polymerization of LBG is about 1,000 (Maier *et al*., 1993; Dea and Morrison, 1975; Srivastava and Kapoor, 2005).

Since LBG solutions at low concentrations are used in food industry as an important thickening and stabilizing agent, their chemical composition and purification, and their impact on

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rheological properties, solubility, dilute solution, and other properties are studied widely. The chemical composition of LBG plays an important role in their rheological properties (Kök, 2007). Chemical composition of commercial LBG is 88% galactomannans, 5% other polysaccharides, 6% protein, and 1% ash (Belitz and Grosch, 1999). Commercial products do not always consist of pure endosperm, but may contain residual hull and germ parts (Wielinga and Maehall, 2000). LBG is purified by precipitation with isopropanol alcohol. The comparison between refined and crud LBG has shown that mannose/galactose ratio increases and ash, protein, and fat content decrease by purification (Kök *et al.*, 1999; Kök, 2007; Bouzouita *et al.*, 2007).

LBG solubility is a function of dissolution temperature. In cold water with 5°C, 50% of LBG polysaccharides have been reported to be soluble and the solubility of LBG increases to about 90% at 85°C. Molecular weight and intrinsic viscosity of dissolved polysaccharides of LBG at 5°C have been reported to be 1,060 kg mol^{-1} and 11.8 dL g^{-1}, respectively, while these parameters could go up to 1,150 kg mol^{-1} and 12.5 dL g^{-1}, by increasing temperature to 65°C (Pollard and Fischer, 2006; Pollard *et al.*, 2007).

According to Sittikijyothin *et al.* (2005), LBG solutions have a Newtonian behavior at low shear rates while at higher shear rates apparent viscosity decreases with increasing shear rate. Other authors have reported the shear thinning behavior for LBG solutions (Bourbon *et al.*, 2010; Wu *et al.*, 2009). The climatic and geographic origin of carob seeds and the cultivation mode influence its chemical and rheological properties (Bouzouita *et al.*, 2007).

The aims of this research were to extract LBG from Iranian carob seeds, purify the extracted LBG, and evaluate the physicochemical and rheological properties of Iranian crude LBG (IcLBG) and Iranian purified LBG (IpLBG) in compression with Sigma LBG (sLBG) as a commercial gum.

MATERIALS AND METHODS

The commercial LBG used in this study was obtained from Sigma Company (Sigma, Germany). Seeds of Iranian carob tree grown wild in Kazeroon region in the Fars Province of Iran, located between 55º, 28´-61º, 20´ longitude and 30º, 21´-38º, 17´ latitude, were collected and used to produce IcLBG. Other chemicals used were of analytical grade unless otherwise mentioned.

Endosperm Extraction and Milling

An aliquot amount of Iranian carob seeds (100 g) was immersed in 800 mL of boiling water at 100°C for 2 hours followed by removing the seeds from hot water. Seed endosperms were then separated manually from the germ and hull. The separated endosperms were dried by a freeze dryer (ZIRBUS, Vaco5, Germany), milled by a disc mill and sifted with 125 μm sieve to obtain Iranian crude LBG (IcLBG) (Dakia *et al.*, 2007; Dakia *et al.*, 2008). The powder was stored in an airtight test tube at room temperature for further experiments.

Purification of IcLBG

The crude LBG was purified according to the procedure of Bouzouita *et al.* (2007) with some modifications. Briefly, LBG, an IcLBG solution (include 1 g LBG 100 ml^{-1} distilled water) was prepared and heated at 80°C in a water bath for 30 minutes. After cooling, isopropanol alcohol was added to the solution (isopropanol to LBG solution, 2:1), mixed thoroughly and left for 120 minutes at room temperature. The white fibrous precipitate formed was then collected by filtering the mixture through a metal sieve (with opening diameter of 1 mm) and washed with isopropanol twice. After drying (in freeze dryer), the precipitate was ground to a fine powder and sieved to obtain IpLBG with particle size of smaller

than 125 μm for further experiments (Bouzouita *et al*., 2007).

Chemical Analysis

Moisture, fat, protein, ash, and fiber contents of the samples were determined according to the AOAC approved methods (Horuitz, 2000). The quantity of total carbohydrate content was estimated as the remainders of the samples after protein, fat, ash, and moisture were accounted for.

Solubility Measurement

Solubility of LBG samples was measured at 25 and 80°C, gravimetrically. Solutions of IcLBG, IpLBG, and sLBG in distilled water (0.1% w/v), were prepared and stirred using a magnet stirrer at ambient temperature (25°C) or 80°C for 30 minutes. Afterwards, the solutions were centrifuged (1800*g*) at room temperature for 30 minutes to remove any insoluble particles. The supernatant of each solution was then separated and dried in an electrical oven (105°C, 24 hours) and dry weight of this fraction was taken as soluble materials and the solubility was calculated using Equation 1 (Dakia *et al*., 2008)

$$\text{Solubility \%} = \frac{\text{Supernatant concentration (mg mL-1)}}{\text{Initial preparation concentration (mg mL-1)}} \times 100$$

(1)

Solution Preparation

LBG solutions were prepared at different concentrations (w/w) by dispersing the required amounts of each gum in distilled water, under slow stirring at room temperature. The dispersions were then heated in a water bath set at 80°C for 15 minutes followed by homogenizing by a homogenizer (IKA T25 Digital, Germany) for 2 minutes at 12,800*g* and stirring by a magnetic stirrer at room temperature for 90 minutes for complete hydration prior to assessment. The pH of LBG solutions

(ranging from 2.5 to 10) was adjusted using immidazol buffer (0.05 g 1 L^{-1} water) as solvent and 0.1M NaOH and/or HCl solutions were used for pH adjustment.

Rheological Measurement

The effects of temperature (5-60°C), pH (2.5-10) and concentration (0.5-1.5% w/v (g mL^{-1}) for IcLBG and 0.375-0.75% w/v (g mL^{-1}) for IpLBG and sLBG on rheological properties were studied. Preliminary experiments showed that viscosities of IpLBG and sLBG were higher than that of IcLBG at the same concentrations and temperatures; therefore, due to torque limit of the viscometer, the measurement of viscosities of IpLBG and sLBG at concentrations above 0.75% was impossible. The viscosity of IcLBG at concentrations below 0.5% was also too low to be measured with suitable precision. Therefore, rheological properties of IpLBG and sLBG were investigated at similar concentrations and IcLBG was studied at higher concentrations.

Rheological behavior of LBG solutions was investigated by a rotational viscometer (Brookfield Viscometer, DVII plus, Brookfield Engineering, USA) equipped with a CP51 (cone and plate) geometry and a temperature control system as a function of hydrocolloid concentration, temperature, and pH in a shear rate range of 0.5-200 s^{-1}.

For each test, 0.5 mL sample was transferred to the sample compartment and, after temperature equilibrium, it was subjected to a programmed shear rate increasing linearly from 0.5 to 200 s^{-1} in 41 steps with interval time of 10 seconds. The flow behavior index and consistency index values were computed by fitting the data to the Power law model using the software Rheocal 32 (Equation (2)):

$$\tau = k\gamma^{\circ n}$$

(2)

Where, τ is the shear stress (mPa), γ° is the shear rate (s^{-1}), k is the consistency coefficient (mPa s) and n is the flow

behavior index (dimensionless) (Garcia-Ochoa and Casas, 1992).

Intrinsic Viscosity

An Ostwald U-tube viscometer (SCHOTT GERÄTE, Germany) submerged in a water bath (Peco, Iran) was used to measure the passage time of the LBG solutions flowing through the capillary. The capillary viscometer was filled with 2 mL of the sample and equilibrated at 50±0.1°C for 5 minutes. Critical concentration of each LBG sample was determined based on the change of slope between dilute and concentrated regions. The viscosities measured were converted to specific viscosity (η_{sp}) using the following equation:

$$\eta_{sp} = \frac{(\eta - \eta_s)}{\eta_s}$$ (3)

Where, η is the viscosity of the solution and η_s is the viscosity of the solvent. According to the definition of intrinsic viscosity ($[\eta]$):

$$[\eta] = \lim_{c \to 0} \frac{\eta_{sp}}{C}$$ (4)

$$\text{or} [\eta] = \lim_{c \to 0} \frac{d \ln \eta_{rel}}{dC}$$ (5)

Where, η_{rel} is the relative viscosity defined as η/η_s and C is the concentration of the solution. Therefore, the experimental data obtained for a set of LBG solutions in a highly dilute region can be analyzed in different ways to obtain intrinsic viscosity. Particularly, in this study, the slope of η_{sp} vs. C plot in the dilute region gave the first estimation for intrinsic viscosity (Chou and Kokini, 1987). Extrapolation to a zero concentration by using Huggins and Kramer rules, which consist of plotting η_{sp}/C vs. C (Equation (4)) and $ln \eta_{rel}/C$ vs. C (Equation (5)), respectively, was used to compare the results obtained from the first technique (Lapasin and Pricl, 1995). The solvent used was 0.1N sodium hydroxide to have the maximum solubility.

Scanning Electron Microscopy (SEM)

SEM images of freeze dries sample was obtained using a scanning electron microscope (Cambridge, UK).

Statistical Analysis

In this research, properties of the Iranian crude and purified LBG and a commercial sample (Sigma LBG) were compared. All measurements were performed in triplicates. Values of all parameters are presented as mean±standard deviation. To compare the data, ANOVA was performed using SPSS 16. The comparison between the means was done using the Duncan multiple range tests with a significance level of $P < 0.05$.

RESULTS AND DISCUSSION

Extraction Yield

The endosperm production from Iranian locust bean seeds, as calculated based on seed weight, was 66.34%, which was higher than the 51-61% yield range reported by Dakia *et al.* (2008). The yield value for the purified LBG was 28.31% based on crude LBG.

Chemical Composition

Table 1 shows the chemical composition of IcLBG, IpLBG, and sLBG samples. As the main constituent of all three LBGs, the total carbohydrate of IcLBG, IpLBG and sLBG was 90.87, 97.96, and 97.22 %, respectively. The protein content of IcLBG (7.13%, dry basis) was significantly greater than that of IpLBG (0.52%) and sLBG (0.58%). Apparently, the purification process reduced the protein content of LBG largely. The results showed that purified LBG (IpLBG) included lower amounts of non-carbohydrate components such as fat and ash. The fiber content of the purified

Table 1. Chemical composition of Iranian crude and purified LBG and Sigma LBG (%, w/w, dry weight basis).

Type	Composition (%)				
	Fat	Protein	Ash	Fiber	Total carbohydrate
Iranian crude LBG	1.05[b][a]± 0.00	7.13[b]± 0.21	0.95[b]± 0.03	2.14[c]± 0.06	90.87[a]± 0.18
Iranian purified LBG	0.97[a]± 0.01	0.52[a]± 0.02	0.55[a]± 0.02	0.74[a]± 0.04	97.96[b]± 0.01
Sigma LBG	1.06[c]± 0.00	0.58[a]± 0.01	1.13[c]± 0.10	0.95[b]± 0.02	97.22[b]± 0.11

[a] Each value is the average of three replicates±standard deviation. In each column, different letters indicate significant difference (P< 0.05).

LBG was lower than the crude LBG, in agreement with the results of Bouzouita *et al*. (2007) and Kök *et al*. (2007).

Solubility of LBG Samples

Table 2 shows solubility values of IcLBG, IpLBG, and sLBG. According to these results, IcLBG is partially cold-water soluble (~ 55% at 25°C) and needs heating to reach a higher solubility value (~ 71% at 80°C). This is in agreement with the findings of Dakia *et al*. (2008), who reported the solubility of carob gum of about 50% at 25°C and 70-85% at 80°C.

At high temperatures, some of the molecules, such as those with higher molecular weight and low galactose content galactomannans, were soluble, while they were insoluble at low temperatures; and these were the causes of increase in solubility with higher temperature (Garcia-Ochoa and Casas, 1992). There was no significant difference between the IpLBG and IcLBG in terms of solubility in hot water. Cold-water solubility of IpLBG (61.53%) was higher than IcLBG and sLBG (54.59 and 54.66%, respectively). It may indicate that during purification, i.e. alcohol precipitation, protein, fiber, and ash contents of IpLBG decrease, therefore, other materials are concentrated.

Rheological Properties

Intrinsic Viscosity

The intrinsic viscosity of a polymer is a measure of the hydrodynamic volume occupied by that macromolecule, which is closely related to the size and conformation of the macromolecular chains in a particular solvent (Lai and Chaing, 2002). Critical concentrations of IcLBG, IpLBG, and sLBG were, respectively, 0.6, 0.25, and 0.15% (w/v). The plots of reduced viscosity and inherent viscosity versus concentration were

Table 2. Solubility (%, w/w) of Iranian crude and purified LBG and Sigma LBG in cold (25°C) and hot (80°C) water.

Type	Solubility (%)	
	Solubility in cold water	Solubility in hot water
Iranian crude LBG	54.59[a][a]± 0.42	71.47 [a]± 1.60
Iranian purified LBG	61.53 [b]± 3.09	71.36[a]±1.18
Sigma LBG	54.66 [a]± 0.30	74.58 [b]± 0.58

[a] Each value is the average of three replicates ± standard deviation. In each column, different letters indicate significant difference (P< 0.05).

used for estimation of intrinsic viscosity (Figures 1).

Intrinsic viscosities of IcLBG, IpLBG, and sLBG samples were 3.95, 9.05, and 9.48 dL g^{-1}, respectively (Table 3). Richardson *et al.* (1998) reported that intrinsic viscosity of LBG is 13.79 dL g^{-1}, while in the reports of Higiro *et al.* (2007), Sittikijyothin *et al.* (2005), and Azero and Andrade (2000) intrinsic viscosity of LBG was calculated as 12.49, 11.03, and 13.3 dL g^{-1}, respectively. This variation could be due to the source of carob seeds, extraction conditions, or solvent type.

Flow Behavior Properties of LBG Solutions

Table 4 shows consistency coefficient (k) and flow behavior index (n) of IcLBG, IpLBG, and sLBG at different concentrations and temperatures. Almost all LBG solutions showed shear thinning flow behavior with flow behavior index of less than one. Figure 4 shows shear stress versus shear rate for 0.75% LBG solutions at 40°C.

Temperature had a positive effect on flow behavior index (except for IcLBG at 0.5%), while flow behavior index was inversely proportional to hydrocolloid concentration. Consistency coefficient decreased with temperature, but increased with concentration. Consistency coefficient of IcLBG was in the range of 11.61 (at 0.5%
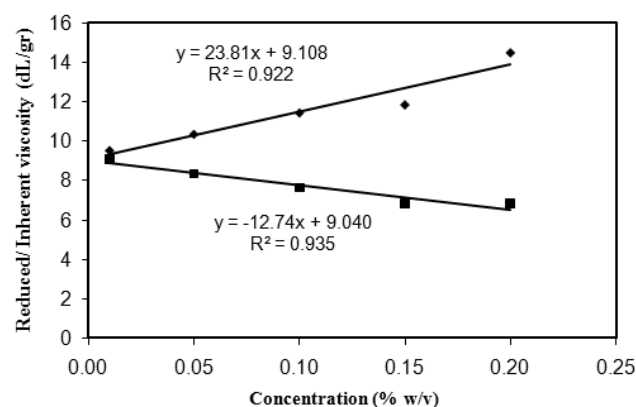
Table 3. The intrinsic viscosity (dL g^{-1}) of Iranian crude and purified LBG and Sigma LBG.

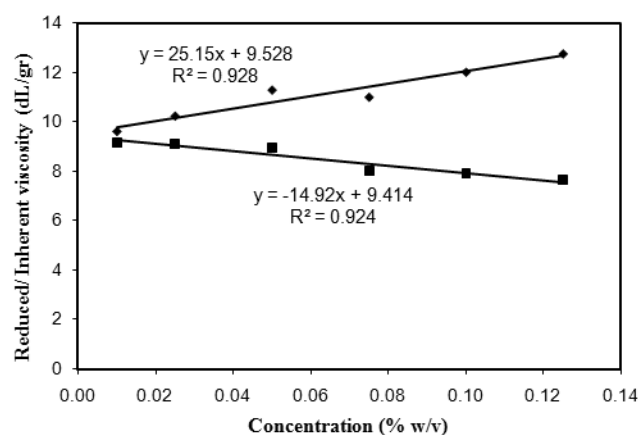| Hydrocolloid	Intrinsic viscosity
Iranian crude LBG	3.95[a][a] ± 0.09
Iranian purified LBG	9.05[b] ± 0.26
Sigma LBG	9.48[c] ± 0.15

[a] Each value is average of three replicates ± standard deviation. Different letters indicate significant difference (P< 0.05).
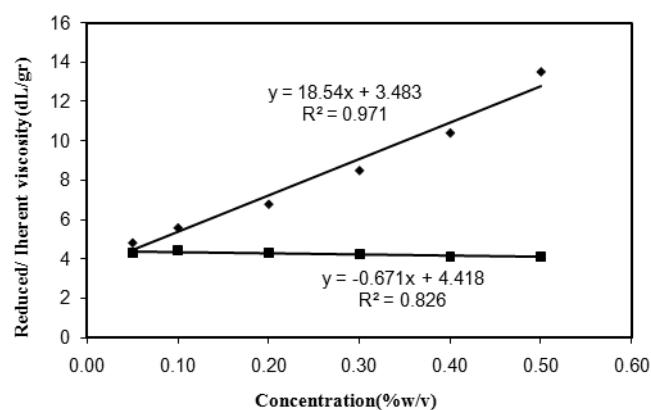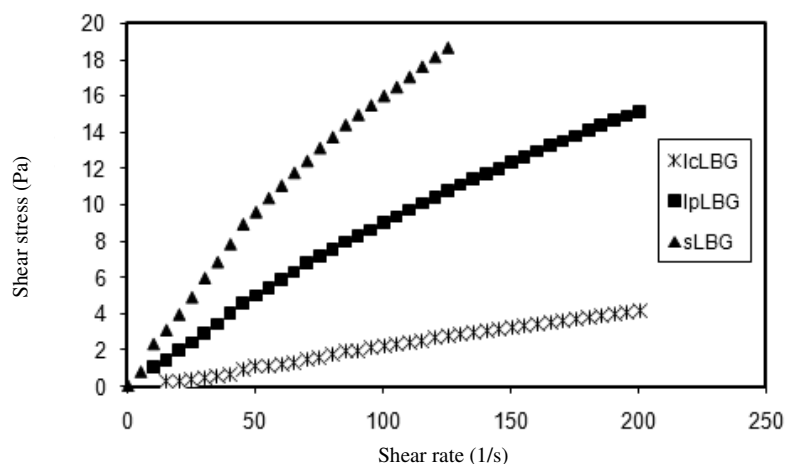

(a)

(b)

(c)

Figure 1. Reduced (♦) and inherent (■) viscosity (dL g^{-1}) versus concentration (% w/v) for (a) IcLBG, (b) IpLBG and (c) sLBG.

130

Figure 4. Shear stress as a function of shear rate for 0.75% (w/v) concentration IcLBG, IpLBG and sLBG at 40°C.

and 60°C) and 2,137.67 mPa s (at 1.5% and 5°C) and flow behavior index values showed that IcLBG solutions were more shear thinning at higher concentrations and lower temperatures.

The shear thinning behavior of LBG solutions has been reported in previous studies (Sittikijyothin *et al*., 2005; Wu *et al*., 2009; Bourbon *et al*., 2010). With increasing shear rate, the molecular entanglements are destroyed such that the molecules align in the direction of flow, and viscosity decreases. However, Newtonian flow behavior of LBG solutions has been reported at very low shear rates or concentrations by some researchers (Dakia *et al*., 2008).

The comparison of rheological data of IcLBG and IpLBG indicated that at the same concentrations and temperatures, IpLBG was more shear thinning with greater consistency coefficient (Table 4). These results are in agreement with the report of Bouzouita *et al*. (2007) in which purified LBG samples showed more shear thinning behavior than their corresponding crude LBG and a significant difference was found between viscosity of purified and crude LBG. They stated that rheological properties of LBG depend upon protein, amount of galactomannan, degree of branching and botanical origin.

Comparison of flow properties of IcLBG, IpLBG, and sLBG (Table 4) indicated that sLBG was able to produce more viscous solutions than IcLBG and IpLBG at the same concentration and temperature. The higher consistency of sLBG solutions than Iranian LBG can be due to the fact that sLBG, as a commercial product, is produced from selected varieties, while in this research carob seeds were collected from carob trees grown wild in a low precipitation region without irrigation. By optimizing the seed collection time of the year and using improved agricultural management and possibly molecular biology techniques, the functional properties of Iranian LBG can be improved.

Effect of pH on Rheological Behavior and SEM images of LBG Solutions

Table 5 shows that, by increasing pH from 2.5 to 10, the consistency coefficient of IcLBG solutions increased significantly. However, consistency coefficient of IpLBG solutions increased up to pH 7, but, by changing pH from 7 to 10, a significant decrease in consistency coefficient was observed. For sLBG, an increase in

Table 4. Consistency coefficient (mPa s) (k) and flow behavior index (n) of Iranian crude and purified LBG and Sigma LBG at different concentrations (% w/v) and temperatures (°C).

Iranian crude LBG

Concentration (% w/v)	0.5		0.75		1		1.5	
Temperature (°C)	k	n	k	n	k	n	k	n
5	$29.60^{b,A}$	$0.96^{b,C\,a}$	$258.10^{d,B}$	$0.74^{a,B}$	$726.17^{d,C}$	$0.65^{a,A}$	nd	nd
25	$25.77^{b,A}$	$0.89^{a,D}$	$99.60^{c,A}$	$0.82^{b,C}$	$350.40^{c,B}$	$0.70^{b,B}$	$2137.67^{c,C}$	$0.62^{a,A}$
40	$14.70^{a,A}$	$0.93^{ab,C}$	$53.57^{b,AB}$	$0.85^{bc,B}$	$122.77^{b,B}$	$0.83^{c,B}$	$1124.67^{b,C}$	$0.67^{b,A}$
60	$11.61^{a,A}$	$0.91^{ab,B}$	$16.40^{a,A}$	$0.89^{c,B}$	$57.10^{a,B}$	$0.94^{d,B}$	$432.83^{a,C}$	$0.80^{c,A}$

Iranian purified LBG

Concentration (% w/v)	0.375		0.5		0.625		0.75	
Temperature (°C)	k	n	k	n	k	n	k	n
5	$48.73^{c,A}$	$0.93^{a,D*}$	$236.70^{d,B}$	$0.73^{a,C}$	$998.13^{d,C}$	$0.62^{a,A}$	$1091.33^{d,D}$	$0.66^{a,B}$
25	$20.07^{b,A}$	$0.99^{b,D}$	$86.77^{c,B}$	$0.83^{b,C}$	$367.97^{c,C}$	$0.72^{b,B}$	$750.70^{c,D}$	$0.65^{a,A}$
40	$7.55^{a,A}$	$1.07^{c,D}$	$45.20^{b,B}$	$0.88^{c,C}$	$129.10^{b,C}$	$0.84^{c,B}$	$462.97^{b,D}$	$0.68^{b,A}$
60	nd	nd	$9.81^{a,A}$	$1.10^{d,C}$	$44.80^{a,B}$	$0.97^{d,B}$	$274.70^{a,C}$	$0.73^{c,A}$

Sigma LBG

Concentration (% w/v)	0.375		0.5		0.625		0.75	
Temperature (°C)	k	n	k	n	k	n	k	n
5	$144.30^{d,A}$	$0.80^{a,C*}$	$2184.33^{c,B}$	$0.45^{a,A}$	$1720.00^{d,B}$	$0.57^{a,B}$	$5050.64^{b,C}$	$0.44^{a,A}$
25	$44.40^{c,A}$	$0.91^{b,D}$	$559.90^{b,B}$	$0.62^{b,B}$	$1313.67^{c,D}$	$0.55^{a,A}$	$1024.67^{a,C}$	$0.68^{d,C}$
40	$18.37^{a,A}$	$1.02^{c,C}$	$141.30^{a,B}$	$0.79^{c,B}$	$798.23^{b,C}$	$0.60^{b,A}$	$1178.33^{a,D}$	$0.60^{b,A}$
60	$25.77^{b,A}$	$0.90^{b,C}$	$123.13^{a,B}$	$0.77^{c,B}$	$199.53^{a,C}$	$0.79^{c,B}$	$714.13^{a,D}$	$0.65^{c,A}$

[a] Each value is the average of three replicates. In each column, different small letters indicate significant difference at different temperatures (P< 0.05). In each row, different capital letters indicate significant difference at different concentrations (P< 0.05). nd: Not determined. In all cases R^2 was greater than

Table 5. Effect of LBG solution pH (2.5-10) on consistency coefficient (mPa s) (k) and flow behavior index (n) of Iranian crude LBG, Iranian purified LBG and Sigma LBG solutions (1% w/v), at 25 ºC.

Hydrocolloid type	Iranian crude LBG		Iranian purified LBG		Sigma LBG	
pH	n	k	n	k	n	k
2.5	$0.81^{d\,a}$	135.50^a	1.01^c	32.60^a	0.62^b	544.50^c
4	0.76^c	217.90^b	0.95^b	53.53^b	0.59^a	642.17^d
7	0.70^b	341.70^c	0.82^a	118.07^c	0.64^c	477.13^b
10	0.69^a	382.97^d	1.01^c	33.77^a	0.65^c	436.03^a

[a] Each value is the average of three replicates±standard deviation. In each column, different letters indicate significant difference at different pH (P< 0.05).

consistency coefficient was seen up to pH 4, but afterwards the consistency coefficient decreased.

The results indicate that viscosity at acidic pH is lower than neutral condition. This is probably due to the deformation of water chemical structure, which influences interaction of water-galactomannan and galactomannan-galactomannan molecules. High concentrations of H^+ in acidic conditions cause weak protonation of hydroxyl groups of water and galactomannan molecules, which results in reducing the number of hydrogen bonding. Under these conditions, less hydrogen bond between water and polymers are formed, which means less expansion and, therefore, fewer interactions of galactomannans causing viscosity reduction (Farahnaky *et al.*, 2013).

Viscosity of IpLBG and sLBG solutions decreased under alkaline condition. Doyle *et al.* (2009) studied galactomannans under alkaline conditions and reported that hydroxyl groups of galactomannan were ionized and polymer molecules became anionic polyelectrolites that caused electrostatic repulsion and, therefore, decrease in viscosity.

Considering the chemical composition of the three LBG samples (Table 1), it was found that the protein content of IcLBG was markedly greater than IpLBG and sLBG and viscosity of IcLBG solutions increased up to pH 10 (the viscosity of IpLBG and sLBG did not show the same trend). This is probably attributed to the presence of proteins rich in acidic amino acids in which

the presence of carboxyl groups retards the ionization of hydroxyl groups of galactomannans. Therefore, electrostatic repulsion between the macromolecules is generated less and viscosity does not decrease.

The microstructure of freeze-dried samples of 1% IcLBG solutions and in a range of pH between 2.5 to 10 was studied using scanning electron microscopy (Figure 5). Overall, the SEM images showed the ability of the Iranian LBG to disperse and form a three dimensional network to entrap water molecules and viscosify the solution. The SEM images confirmed the results obtained by viscometry method.

CONCLUSIONS

In this study, locust bean gum was extracted from Iranian carob seeds with yield of 62.34% and it was then purified successfully. Physicochemical and rheological properties of the Iranian crude and purified LBGs were measured and compared with a commercial LBG. Chemical compositions of the LBG samples showed that protein contents of IcLBG and sLBG were 7.13 and 0.58%, respectively. This difference could be regarded as one of the reasons behind differences of rheological properties. The solubility data showed that heating step could play a major role in solubilizing all three LBG samples. All three samples showed shear thinning behavior and were able to thicken aqueous solutions; moreover, sLBG had the greatest

(a)

(b)

(c)

(d)
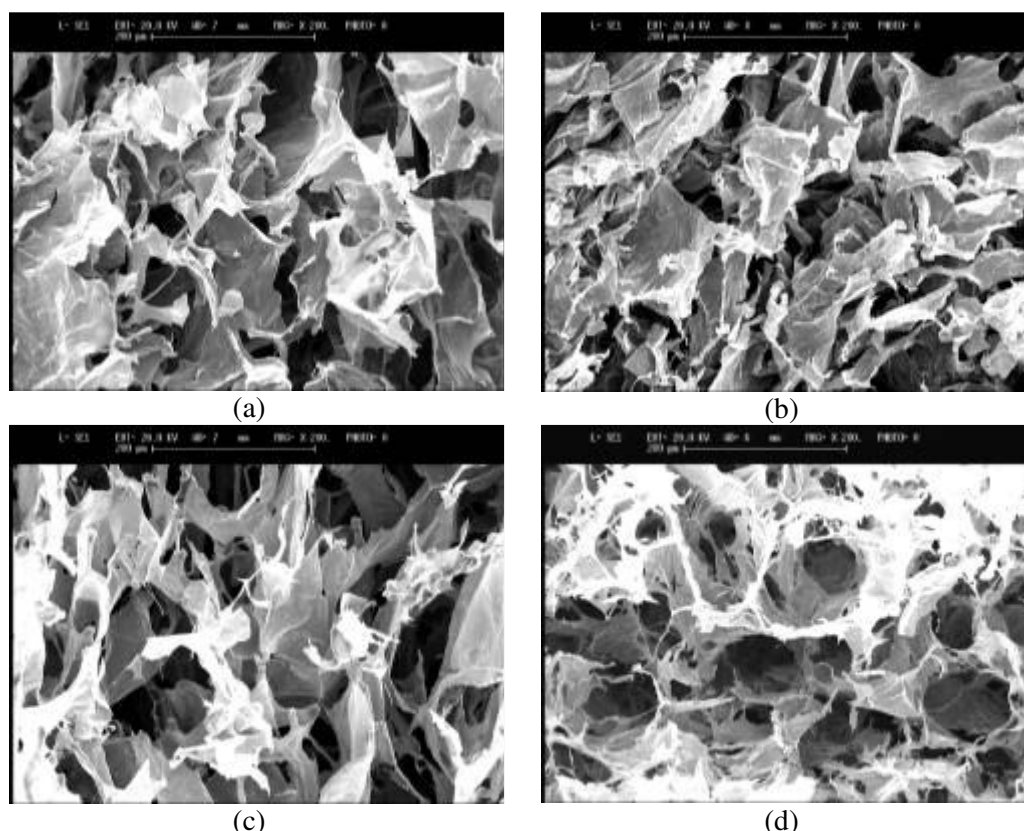
Figure 5. SEM Images (×200) of freeze dried samples of IcLBG solutions (1% w/v) A: pH= 2.5; B: pH= 4;   C: pH= 7 , D: pH= 10.

consistency coefficient. Taken together, it can be concluded that the Iranian carob seed is a suitable source for production of LBG gum, however, more research is needed to improve the functional properties of the extracted Iranian LBG gum using different approaches.

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خواص فیزیکوشیمیایی صمغ دانه خرنوب معمولی و خالص استخراج شده از دانه های خرنوب ایرانی

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چکیده

در این پژوهش، دانه های درختان خرنوب، که به صورت خودرو در شهر کازرون (استان فارس) رشد نموده بودند برای تهیه صمغ دانه خرنوب محلی ایران، مورد استفاده قرار گرفت. به منظور مقایسه نتایج، از یک نمونه صمغ دانه خرنوب تجاری (سیگما) استفاده شد. ترکیبات شیمیایی، حلالیت و خواص رئولوژیکی نمونه های صمغ مورد بررسی قرار گرفت. نتایج نشان داد میزان پروتئین در صمغ دانه خرنوب محلی ایران (خام) بیشترین مقدار بود و ترکیبات شیمیایی صمغ دانه خرنوب خالص محلی ایران و صمغ دانه خرنوب تجاری مشابه بودند. در بررسی میزان حلالیت مشخص شد که صمغ دانه خرنوب محلی ایران (خام) در آب سرد حلالیت نسبی داشته (۵۵٪ در $25\,^\circ C$) و برای افزایش حلالیت باید دما را افزایش داد (۷۱٪ در $80\,^\circ C$). خواص رئولوژیکی محلول های هر سه نمونه صمغ به عنوان تابعی از غلظت و دما ($60\,^\circ C - 5\,^\circ C$) در محدوده تنش برشی (۱/S)۲۰۰–۰/۵ مورد بررسی قرار گرفت. هر سه نمونه، رفتار رقیق شونده با برش داشته و رفتار رئولوژیکی آن ها با مدل قانون توان مطابقت داشت. نتایج نشان داد صمغ دانه خرنوب محلی ایران، توانایی ایجاد قوام در محلول های آبی را دارد، با این وجود قدرت قوام دهندگی آن از نمونه تجاری کمتر بود. بررسی ها نشان داد pH (۱۰–۲/۵) به صورت معنی داری بر خواص رئولوژیکی محلول های صمغ تاثیر دارد. ویسکوزیته ذاتی صمغ دانه خرنوب خام محلی ایران، صمغ دانه خرنوب خالص محلی ایران و صمغ دانه خرنوب تجاری به ترتیب ۳/۹۵، ۹/۰۵ و ۹/۴۸ (dL/g) محاسبه شد.

136