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Physical and rheological characterization of seeds of three legume trees

Caracterización física y reológica de semillas de tres leguminosas arbóreas

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RESUMEN

El algarrobo chileno (*Prosopis chilensis* (Mol) Stuntz), la tara (*Caesalpinea spinosa* (Mol) Kuntze) y la acacia de las tres espinas (*Gleditsia triacanthos* L) ofrecen un interesante potencial para promover el desarrollo de las zonas áridas. En el endosperma de sus semillas existe una goma del tipo galactomanano que posee propiedades viscosantes y estabilizadoras de emulsiones. Se realizó una investigación para evaluar las características físicas de las vainas y semillas y las propiedades reológicas de las gomas de estas especies. Se estudiaron frutos de cinco ecotipos de algarrobo (Región de Coquimbo y Región Metropolitana), de tara (Región de Coquimbo) y de acacia de las tres espinas (Región Metropolitana). Se midieron las dimensiones de las vainas, número de semillas/vaina, peso de semillas, rendimiento y parámetros de color de cotiledón y goma y las propiedades reológicas de las gomas extraídas. Las gomas se extrajeron manualmente y se secaron a 50 ºC. En algarrobo, las semillas pesaron 0,04 g y el rendimiento de goma varió entre 25,5 y 31,5%. Las semillas de tara pesaron 0,14 g y las de acacia de las tres espinas 0,16 g, con un rendimiento de goma 31,0 y de 40,1% y respectivamente. Las gomas presentaron una alta luminosidad y contribución del amarillo y baja contribución de rojo. Las dispersiones, en todas las gomas estudiadas, presentaron un comportamiento no newtoniano del tipo Herschel-Bulkley, correspondiendo a un fluido inelástico, independiente del tiempo. La viscosidad aparente de la goma de algarrobo fluctuó entre 0,020 y 0,12, la de tara entre 0,06 y 1,3, y la de acacia de las tres espinas entre 0,034 y 0,08 Pa*s. Las gomas de estas especies tienen características reológicas que permitirían su aplicación en alimentos.

Palabras clave: algarrobo, tara, acacia de las tres espinas, hidrocoloides, galactomananos.

SUMMARY

*The Chilean algarrobo (*Prosopis chilensis *(Mol) Stuntz), the tara (*Caesalpinea spinosa *(Mol) Kuntze) and the honey locust (*Gleditsia triacanthos *L) have an interesting potential for the development of arid zones. The endosperm of the seeds of these species has a galactomannan-type gum which has the property of thickening and stabilizing emulsions. This study evaluates the physical characteristics of the pods and seeds, and the rheological properties of the gums of these species. We studied fruits of five ecotypes of algarrobo (Coquimbo and Metropolitan Regions), of tara from the Coquimbo Region and of honey locust (three-spined acacia) from the Metropolitan Region. We measured the dimensions of the pods, number of seeds per pod, seed weight, gum yield and color parameters of the cotyledon and gum, and the rheological properties of the gums extracted. Gums were extracted manually and dried at 50º C. The seeds of algarrobo weighed 0.04 g; their gum yield varied between 25.5% and 31.5%. The seeds of tara weighed 0.14 g and those of the acacia 0.16 g, with a gum yield of 31.0% and 40.1%, respectively. The gums had high luminosity and high contribution of yellow, along with low contribution of red. The dispersions of all the studied gums showed a non-Newtonian Herschel-Bulkley behavior corresponding to an inelastic fluid, independent of time. The apparent viscosity of the gum of algarrobo fluctuated between 0.020 and 0.12; that of tara between 0.06 and 1.3, and that of honey locust between 0.034 and 0.08 Pa*s. The gums of these species have rheological characteristics which would allow their addition to foods.*

Key words: *algarrobo, tara, honey locust, hydrocolloids, galactomannans.*

Introduction

In recent years have realized that in order to preserve the environment, must maintain biodiversity and re-evaluate native species, especially trees, which may be used to obtain food and products of industrial use (Ramachandran, 2007). This has produced initiatives to consider underutilized plants, especially in areas in which basic crops are hard to establish due to salinity or lack of water (Barba de la Rosa *et al.*, 2006). Among these, the Chilean algarrobo (*Prosopis chilensis*

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(Mol) Stuntz) has an interesting potential for the development of arid zones. Two related species, the tara (*Caesalpinea spinosa* (Mol) Kuntze) and the honey locust (*Gleditsia triacanthos* L) have been little exploited in Chile.

Many legume seeds develop an endosperm composed of polysaccharides which serve as an energy and water reserve (Pollard *et al.*, 2010). These seeds are composed of a seed coat, endosperm and cotyledons in different proportions.

There are a number of ecotypes of algarrobo in Chile which have different fruit and seed characteristics. Fruits are indehiscent pods with 17-21 seeds per fruit. Their composition is: seed coat, 13%-19%; gums (endosperm), 28%-31%; and cotyledon, 39%-43% (Escobar *et al.*, 1987). Escobar *et al.* (2005) reported that the fruit of the honey locust has an average length of 36.7 cm and 22 seeds per fruit; seeds are dark brown, very hard and smooth with average diameter of 0.75 cm. For tara, Estévez *et al.* (2006) indicated that fruit length varied from 5.5-8.3 cm, with 5-6 seeds per fruit; seeds had a diameter of 0.65 cm and were similar to those of the honey locust.

The extraction of the cotyledons and gum from the seeds of legume trees is a slow, difficult process, due principally to the hardness of the seed coat. In algarrobo, tara, honey locust and other species different mechanical, physical and chemical methods have been studied for the removal of the seed coat and for separating the coat, the endosperm (gum) and the cotyledons (Wielinga, 2000; Cerqueira *et al.*, 2009). Chemical extraction methods studied include sodium hydroxide 0.75% w/v and 72% w/v sulfuric acid with different temperatures and contact times (Escobar *et al.*, 1987; Vásquez *et al.*, 1988, Cruz, 1999; Estévez *et al.*, 2004; Vega, 2006 and Martínez, 2007). These methods cause environmental contamination, thus it is important to look for other, less aggressive methods to perform the extraction.

The endosperm of these seeds is composed of galactomannans, which are neutral polysaccharides formed by a linear chain of mannose with lateral substitutions of galactose in different proportions; their properties serve to thicken and stabilize emulsions (Wielinga, 2000; Azero y Andrade, 2002). The relation between mannose and galactose affects the dispersability and the rheological behavior of the galactomannans; a greater quantity of mannose produces greater difficulty of dispersion due to the greater cohesion of the main chain. Thus for example, guar gum, which has high galactose content, disperses easily in cold water, while locust gum requires higher temperature and agitation to achieve an adequate dispersion (Wielinga, 2000; Picout *et al.*, 2002).

The gums of algarrobo, tara and honey locust are galactomannan types of hydrocolloids with different mannose:galactose proportions (M:G), which vary from 1.4:1 in algarrobo to 3.0:1 in tara and from 2.5 to 3.0:1 in honey locust (Presle *et al.*, 2004; Sciarini *et al.*, 2009; FAO, 1995; Picout *et al.*, 2002). Hydrocolloids are used in foods due to their texturizing properties, which play an important role in the acceptability of products. A number of variables, such as the shear rate, temperature, pressure and shear time affect the viscosity of hydrocolloids. For non-Newtonian fluids, at constant temperature and pressure viscosity decreases with an increase in deformation, generating pseudo-plasticity or greater fluidity behavior, depending on the shear (Marcotte *et al.*, 2001).

Based on the above information, this study evaluated the physical characteristics of the pods and seeds of algarrobo, tara and honey locust, and the rheological properties of their gums.

Materials and Methods

We studied the fruits and seeds of five ecotypes of algarrobo, four (A-1, A-2; A-3 y A-4) from the Región de Coquimbo and one (A-5) from the Región Metropolitana of Chile; of tara, collected in the Región de Coquimbo, and of honey locust from the Región Metropolitana. For 20 pods of each ecotype and species we measured dimensions, number of seeds per pod and seed weight. In algarrobo, the gum was extracted after heating in water at 97º C for 5 min and soaking in distilled water 1:3 for 24 hours. For tara and honey locust seeds were soaked in a EDTA 0.25% w/v for 24 hours and then boiled in distilled water for 30 min, after which they were soaked as with algarrobo. Seed coat, endosperm (gum) and cotyledon were separated manually and then dried at 50º C. We determined yield and the color of the cotyledon and gum in a Minolta CR-200b colorimeter.

Dispersions of 0.25% and 0.50% w/v were prepared for the three species to determine their rheological properties at 25º C. Measurement time was 15 min; we measured 150 points for each solution, beginning with a velocity gradient (D) of 0 and ending with 500 s^{-1}.

For each dispersion we measured the apparent viscosity (Pa*s), the flow threshold (Pa) and the index of flow behavior (dimensionless) of the gums extracted, in a Brookfield™ RHEO 2000 R/S rheometer with concentric cylinders, using a CC45 DIN spindle. We plotted flow curves and the viscosity profile for each dispersion.

Results and Discussion

Physical characteristics of the seeds of algarrobo, honey locust and tara

The pods of algarrobo were golden yellow to reddish-yellow, flat, curved, semi-curved and straight. Their length varied from 9.8 to 12.2 cm (Table 1); the ecotype of the Metropolitan Region (A5) was not as long as the values reported by Escobar *et al.* (1987) from the Metropolitan and Coquimbo regions, and by Pasiecznik *et al.* (2004) for species in different parts of the world. The width ranged between 1.0 and 1.4 cm, similar in all ecotypes and within the values reported by Pasiecznik *et al.* (2004). The mean number of seeds per pod ranged from 11.1 to 15.8; those of the Metropolitan Region had the lowest number; these values are lower than those reported by Escobar *et al.* (1987) and by Pasiecznik *et al.* (2004). The yield of seeds in relation to pod varied from 10.8% to 19.2%, similar to the values reported by Figueiredo (1990) and lower than those found by Escobar *et al.* (1987). The gum yield as a proportion of seed weight varied from 25.5% (A-5) to 31.5% (A-2), and that of the cotyledon from 37.8% (A-1) to 40.5% (A-3 and A-4), very similar to the values reported by Escobar *et al.* (1987).

The dimensions of the pods of tara were similar to those obtained by Estévez *et al.* (2006) and somewhat smaller than those reported in FAO (1995). The pods of the honey locust had dimensions similar to those reported by Escobar *et al.* (2005). The seeds of tara and honey locust weighed 0.14 and 0.16 g, respectively, with a yield in seed coat, gum and cotyledon of 41.8%, 31.0% and 26.7% for tara and 25.3%, 40.1% and 28.2% for honey locust. FAO (1995) reported yield values for tara of 38-40%, 22-27% and 26-40% for these fractions. The yield obtained in honey locust was similar to that obtained by Cerqueira *et al.* (2009) in gum extracted with ethanol, and somewhat superior to that reported by Sciarini *et al.* (2009) for purified gum.

The color parameters of the gums of algarrobo were similar among the ecotypes; L* varied from 79.1 to 82.5 (high luminosity; a* from –2.6 to –3.9 (low contribution of red); and b* ranged from 26.8 to 36.2 (high contribution of yellow). These results differ slightly from those reported by Estévez *et al.* (2004) for gums extracted with 0.75% NaOH w/v and 72% H_2SO_4 w/v; these authors found lower luminosity with more participation of green and less participation of yellow. The values of the parameters L*, a* and b* were very similar for tara and honey locust (Table 2). In the honey locust, Martínez (2007), using and NaOH for extraction in the same concentrations as above, found less luminosity of the gum and similar participation of red and yellow.

The cotyledons of algarrobo (Table 3) produced values of L* between 70.7 and 76.4 and low

Table 1. Pod dimensions and extraction yield of seed coats, gums and cotyledons of seeds of algarrobo, tara and honey locust.

Seeds	Pod length (cm)	Pod width (cm)	Number of seeds/pod	Seed weight (g)	Seed coat yield (%)	Gum yield (%)	Cotyledon yield (%)
Algarrobo							
A-1	11.2±2.1[1]	1.4±0.2	14.3±3.5	0.03±0.0	33.9±2.9	28.4±2.5	37.8±3.2
A-2	12.0±1.7	1.0±0.1	15.8±3.7	0.04±0.0	29.9±2.5	31.5±2.8	38.7±3.4
A-3	11.1±1.0	1.0±0.1	13.2±4.3	0.04±0.0	30.0±2.7	29.5±2.6	40.5±3.5
A-4	12.2±1.4	1.3±0.1	12.5±5.7	0.04±0.0	31.7±2.8	27.8±2.5	40.5±3.6
A-5	9.8±1.6	1.3±0.2	11.1±0.6	0.04±0.0	34.9±3.0	25.5±2.4	39.6±3.3
Tara	7.3±1.1	1.7±0.2	3.4±1.0	0.14±0.0	41.8±3.4	31.0±2.7	26.7±2.4
Honey locust	36.9±3.7	3.4±0.2	23.4±2.6	0.16±0.1	25.3±2.3	40.1±3.3	28.2±2.5

[1] Mean ± SD.

Table 2. Color parameters of the gums of seeds of algarrobo, tara and honey locust.

Seeds	L*	a*	b*	H*	C*
Algarrobo					
A-1	82.3±2.9[1]	–3.4±0.8	36.2±1.7	95.4±1.8	36.4±1.7
A-2	80.5±2.7	–3.9±0.6	30.3±1.6	97.3±2.1	30.5±1.5
A-3	82.5±2.4	–3.0±0.4	27.2±1.4	96.2±1.9	27.0±1.6
A-4	79.1±2.0	–3.5±0.7	26.8±1.5	97.5±2.2	27.0±1.4
A-5	81.7±2.3	–2.6±0.5	28.6±1.4	95.2±1.8	28.7±1.5
Tara	74.3±3.2	–0.8±0.6	25.0±0.3	90.9±2.6	25.0±0.2
Honey locust	74.6±5.4	–1.4±0.2	22.8±2.9	93.3±0.1	23.7±2.9

[1] Mean ± SD.

Table 3. Color parameters of the cotyledons of seeds of algarrobo, tara and honey locust.

Seeds	L*	a*	b*	H*	C*
Algarrobo					
A-1	72.8±3.2[1]	1.5±0,1	46.0±0.8	88.1±2.6	46.0±0.7
A-2	71.5±3.0	–1.5±0,0	35.9±0.6	92.4±1.9	35.9±0.5
A-3	76.4±3.5	–0.6±0,0	42.2±0.7	90.8±2.4	42.2±0.6
A-4	70.7±2.7	0.1±0,0	38.3±0.9	89.9±2.5	38.3±0.8
A-5	71.6±2.9	0.4±0,0	38.3±0.5	89.4±2.7	38.3±0.7
Tara	75.8±3.5	–0.5±0,1	28.9±0.3	91.0±3.0	28.9±0.4
Honey locust	79.7±1.3	1.8±0,1	33.8±1.6	87.0±1.5	33.8±1.3

[1] Mean ± SD.

participation of green, very similar to the results of Escobar *et al.* (2009); there was a very high participation of yellow (35.9-46.0), which indicates a more yellow color than that reported by Escobar *et al.* (2009) for cotyledons extracted with NaOH (29.1). The cotyledons of honey locust had an L* value of 79.7 and b* of 33.8, similar to the values reported by Escobar *et al.* (2005), however, the a* value was much lower than those they reported; the color of the cotyledons is an intense yellow. Castillo (2008) indicated quite different color parameters in cotyledons extracted with H_2SO_4, which had greater luminosity and less contribution of yellow. The L* value of tara indicated less luminosity than that obtained by Estévez *et al.* (2006), with a similar participation of yellow and less contribution of red, indicating a clear yellow color.

Rheological characteristics of the gums of algarrobo, honey locust and tara

The dispersions of the gums, in all the ecotypes of algarrobo analyzed, showed a non-Newtonian Herschel-Bulkley behavior corresponding to an inelastic fluid, independent of time with $R^2 = 0.999$. Vásquez *et al.* (1988) found that dispersions of algarrobo had a pseudo-plastic behavior, intermediate between those of guar and locust gums. Figures 1 and 2 show that there were differences between ecotypes, both in the viscosity curves and in the rheological parameters of the gums; A-1 had the greatest viscosity at 0.25% w/v while A-4 had the greatest value at 0.50% w/v. The viscosity of gums decreased more notably at the highest concentration than in more diluted dispersions, which is in accordance with published information for other hydrocolloids, due to the formation of aggregates of polymers in suspension which are stable at the deformation velocities but are easily broken when the agitation of the system increases (Yaşar, Kahyaoglu & Şahan, 2009).

Rheologically, the dispersions of tara and honey locust showed the same behavior as the gums of algarrobo, with $R^2 = 0.999$ in all cases (Figure 3). Both species had greater viscosity at higher concentrations, with loss of viscosity with an increase in

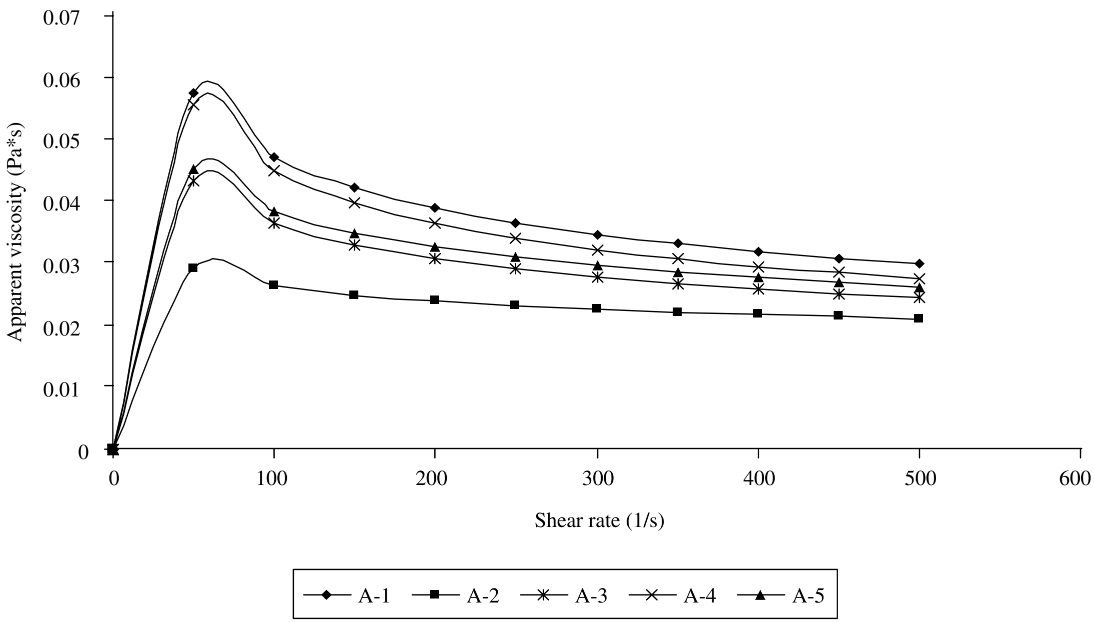

Figure 1. Apparent viscosity curves for gum dispersions of algarrobo at 0.25% w/v.

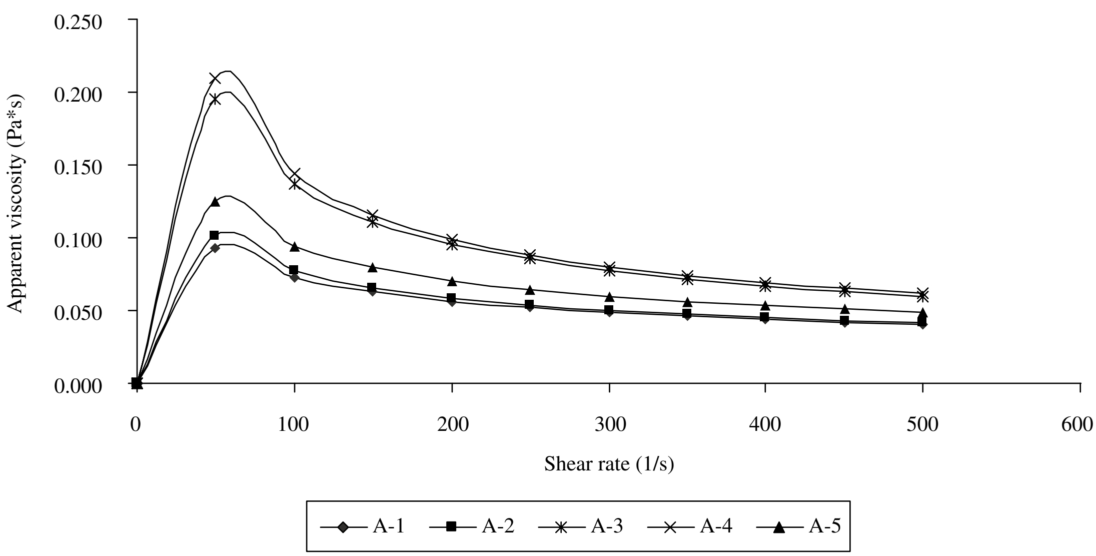

Figure 2. Apparent viscosity curves for gum dispersions of algarrobo at 0.50% w/v

the deformation gradient, as observed for algarrobo. The gum of tara had greater viscosity (beginning of pseudo-plasticity) at a given concentration than that of honey locust and the majority of the algarrobo ecotypes (A-1, A-2 and A-5), which may be related to its low galactose content (Picout *et al.*, 2002). The greater apparent viscosity of honey locust was slightly lower than that reported by Sciarini *et al.* (2009) in alcohol-purified gum.

When the shear rate increased, the shear stress increased for all the dispersions of algarrobo gums (Figures 4 and 5). According to Marcotte *et al.* (2001), as the shear force increases the internal structure of the fluid breaks down, generating an

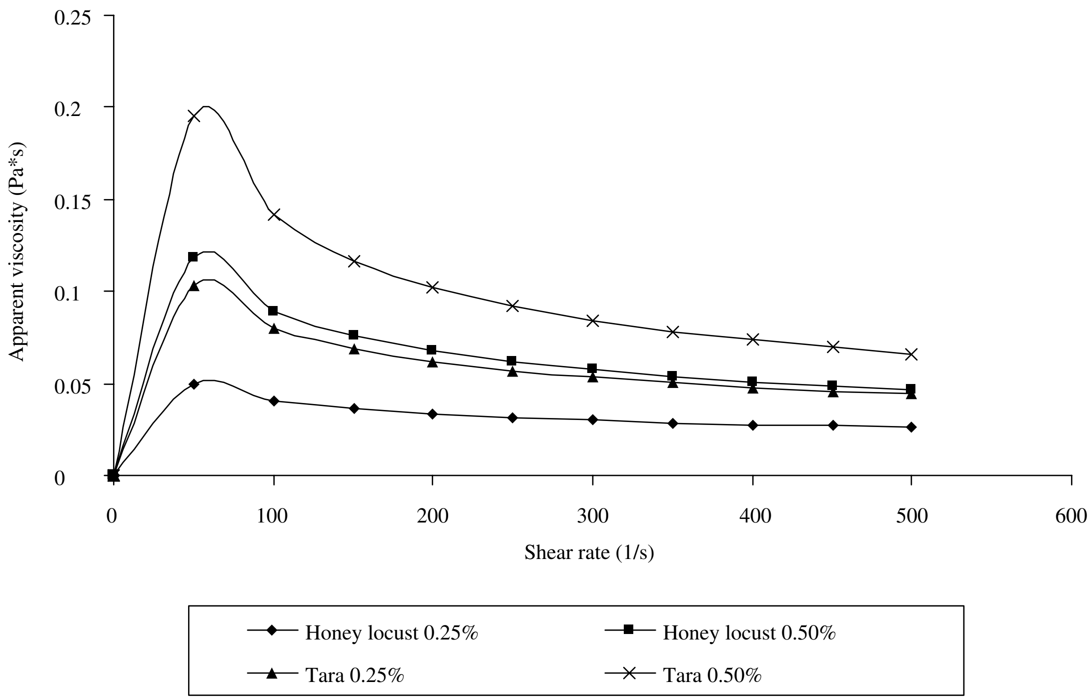

Figure 3. Apparent viscosity curve for gum dispersions of honey locust at 0.25% and 0.50% w/v.

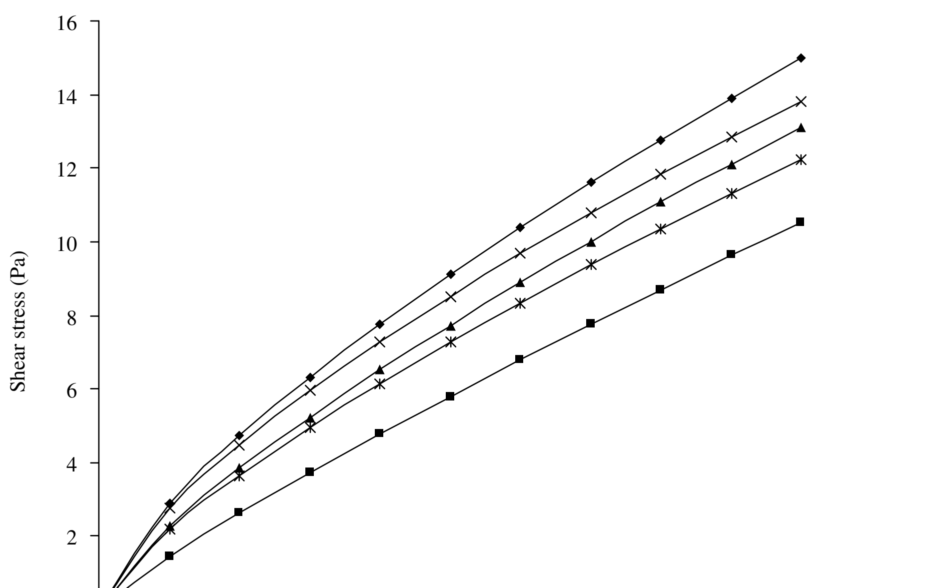

Figure 4. Dispersion flow curve of algarrobo gums at 0.25% w/v.

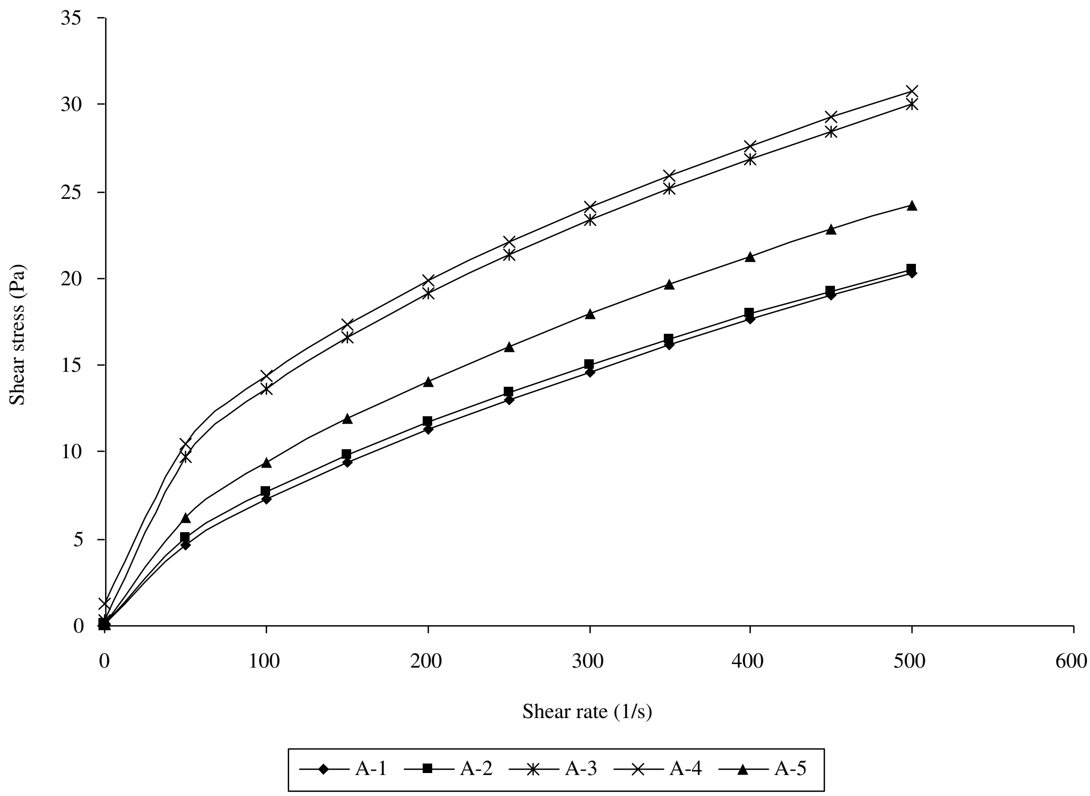

Figure 5. Dispersion flow curve of algarrobo gums at 0.50% w/v.

increase in the flow of the dispersion. The gum from algarrobo ecotype A-2 at 0.25% w/v required more shear stress to reach the maximum deformation, which makes it less stable. However, at 0.50% the behavior of the gums from A-1 and A-2 was very similar. At the higher concentration more shear force was required to reach the same deformation gradient. The gum from A-4 had the most stable behavior under deformation and the greatest viscosity at both concentrations.

The gums of honey locust and tara showed the same tendency as the algarrobo gums (Figure 6); at both concentrations the dispersions of honey locust were less stable than those of tara, and also had lower viscosity. The gum of tara showed the greatest viscosity and stability of all studied gums, which is in agreement with its greater mannose content (Lazaridou *et al.*, 2001).

The flow threshold is the shear force necessary to make a dispersion start to flow. As Table 4 shows, none of the algarrobo guns had a flow threshold (τ_0) at the lowest concentration except for ecotype A-4; in the rest of the gums this parameter only appeared at 0.50% w/v and was greatest in the gum of ecotype A-4, indicating that its flow requires more shear force. The gums of tara and honey locust did not have a flow threshold, although the latter species had a very low value at 0.50% w/v.

In all the dispersions of the studied gums, the consistency index increased with concentration, indicating that they became more viscous (Table 4); tara gum had the highest index value at both concentrations.

The index of flow behavior (n) decreased with increased concentration in all the dispersions, increasing their pseudo-plasticity, agreeing with the results of Vásquez *et al.* (1988) and Marcotte *et al.* (2001) for gum dispersions of algarrobo and commercial hydrocolloids, respectively. In general, at greater concentrations the index of consistency (K) increased and the index of flow behavior decreased, increasing the pseudo-plasticity, in agreement with the results of Dakia *et al.* (2008) for dispersions of locust gum.

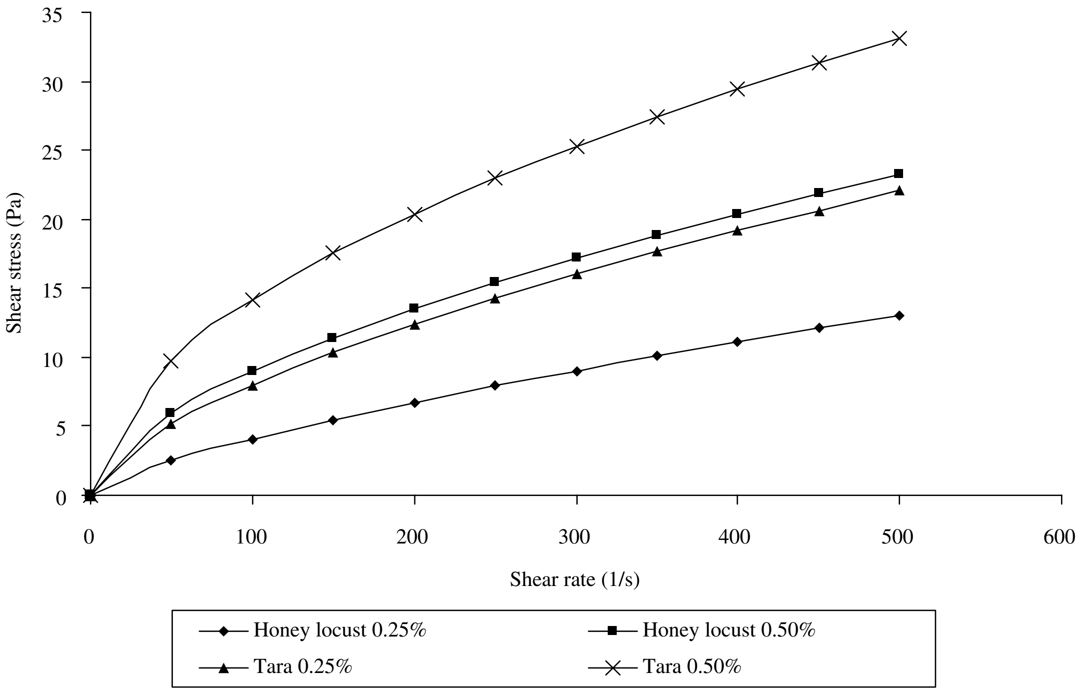

Figure 6. Dispersion flow curves of honey locust gum at 0.25% and 0.50% w/v.

Table 4. Rheological parameters of the Herschel-Bulkley equation for the gums extracted.

Seeds	Flow Threshold τ_0 (Pa)	Consistency Index τ ($Pa^{*}s^{n}$)	Flow Index n
Algarrobo			
A-1 0.25%	0.0	0.17	0.72
A-2 0.25%	0.0	0.05	0.86
A-3 0.25%	0.0	0.12	0.75
A-4 0.25%	0.02	0.18	0.70
A-5 0.25%	0.0	0.11	0.76
A-1 0.5%	0.07	0.37	0.64
A-2 0.5%	0.08	0.45	0.62
A-3 0.5%	0.29	1.35	0.50
A-4 0.5%	1.29	1.27	0.51
A-5 0.5%	0.08	0.60	0.59
Tara 0.25%	0.0	0.44	0.63
Tara 0.5%	0.0	1.23	0.53
Honey locust 0.25%	0.0	0.15	0.72
Honey locust 0.50%	0.03	0.57	0.60

In conclusion, the dispersions of all studied gums showed a non-Newtonian rheological behavior which adjusted to the Herschel-Bulkley model, corresponding to inelastic fluids independent of time, without flow threshold at low concentrations. In all cases the consistency index increased and the flow index decreased at greater concentration. All the studied gums have characteristics which would allow them to be used as texturing agents in foods.

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