



The effect of pH and salt on the stability and physicochemical properties of oil-in-water emulsions prepared with gum tragacanth



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ABSTRACT

The effect of pH (2.5, 4.0 and 5.4) and salt concentration (0.0, 0.5 and 1.0 wt%) on the physical stability of oil-in-water emulsions stabilized with gum tragacanth were investigated during 150 days of storage. Mean droplet diameter, zeta-potential, interfacial tension and steady-shear and dynamic rheological properties were determined to achieve more information about the likely stability mechanisms. The results showed that increasing salt concentration did not have a significant effect on emulsion stability. Emulsions were highly unstable at pH 2.5, with their emulsion-stability index declining almost three times more than that of other emulsions during the storage time. Based on the size distribution data, a direct correlation was not observed between droplet size distribution and emulsion stability. Rheological analysis revealed that pH 2.5 had the lowest apparent viscosity, storage modulus, energy of cohesion (EC) and α -value, and the highest $\tan \delta$ and b -value.

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1. Introduction

An emulsion is a heterogeneous system of two immiscible materials, usually liquids (generally oil and water), where one of the phases (the dispersed phase) is distributed in the other as one continuous phase (Binks, 1998; Dalgleish, 2003). By this wider definition, a considerable number of foods contain emulsions as part of their structure or have been prepared as emulsions at some stage of their processing; examples include mayonnaise, cream and some sauces, desserts, comminuted meat products and beverages (Dickinson & Stainsby, 1982; Herrera, 2012; McClements, 2004). Most emulsions are not thermodynamically stable and tend to break down during storage, through a number of mechanisms (creaming or sedimentation, flocculation, Ostwald ripening, coalescence and phase inversion), yet emulsion stability is one of the most

important factors in managing the shelf-life of foods. An emulsion's stability basically refers to its ability to resist change in its appearance over time. Therefore, it is desirable to make emulsions that are kinetically stable (metastable) for weeks, months or years. Most metastable emulsions that will be encountered contain oil, water and an emulsifying agent, also called an emulsifier or stabilizer.

Emulsifiers can help improve foods' long-term stability by lowering their oil–water interfacial tension and by forming a protective film around the foods' fat droplets to keep them from aggregation. Stabilization is accomplished by increasing the viscosity of the continuous phase, as well as the steric hindrance and electrostatic interactions (Dickinson & McClements, 1996; Dalgleish, 2003; McClements, 2004; Tadros, 2013).

Gum tragacanth consists of dried exudates obtained from the stems and branches of Asiatic species of *Astragalus*; it has a molecular mass of about 840 kDa (Anderson & Bridgeman, 1985; Chenlo, Moreira, & Silva, 2010; Mohammadifar, Musavi, Kiumarsi, & Williams, 2006). It is the best-known hydrocolloid for use in acidic oil-in-water emulsions (Weiping, 2000). Previous studies have reported that gum tragacanth has emulsifying and stabilizing properties, which means that this gum can be suitable for creating stable oil-in-water emulsions (Balaghi, Mohammadifar, &

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Zargaraan, 2010; Dickinson, 2009). Weiping et al. reported that gum tragacanth consists of at least two fractions: a water-swellable constituent known as bassorin and a water-soluble constituent known as tragacanthin (Weiping, 2000). Also, it has been demonstrated that tragacanth-gum samples derived from different species of *Astragalus* have different compositions, and that the ratio of the soluble and insoluble gum fractions strongly varies between species. Therefore, use of these various species of gums as stabilizers, thickeners and emulsifying and suspending agents depends on their rheological properties and composition (Balaghi et al., 2010).

Farzi, Emam-djomeh, and Mohammadifar (2013) studied the emulsifying properties of three species of Iranian *Astragalus*. They concluded that *Astragalus gossypinus* produced the most stable emulsions, although it had lower viscosity than *Astragalus compactus*; they attributed this to the former's higher insoluble fraction and higher uronic content.

Gavlighi et al. reported that the type of gum tragacanth (as determined by the ratio of tragacanthin and bassorin fractions, monosaccharide composition, methoxylation degree and galacturonic acid content) and emulsions-consistency index have a direct effect on stabilizing emulsions in the presence of whey protein isolate. They concluded that the most stable emulsions were achieved with the *Astragalus flucosus* gum incorporated with whey protein isolate. *A. flucosus* had the highest ratio of tragacanthin and bassorin fractions, and emulsions prepared with this gum had the highest consistency index (Gavlighi, Meyer, Zaidel, Mohammadifar, & Mikkelsen, 2013). It has also been shown that gum tragacanth alone, in the absence of protein, could be used to stabilize oil-in-water emulsions. Karimi and Mohammadifar (2014) studied the influence of *A. gossypinus* and *A. flucosus* on the stability of oil-in-water emulsion, finding that *A. gossypinus* had a stronger emulsion-stabilization effect than *A. flucosus*; this property was related to the differences in their tragacanthin/bassorin ratios and chemical composition. Thus, according to previous studies of gum tragacanth's ability to reduce the interfacial tension of the oil-in-water system, the current study examined the use only of *A. gossypinus*, a polysaccharide with low protein content, for stabilizing oil-in-water emulsions. Due to a lack information about the effects of exogenous and endogenous factors on this anionic gum's ability to stabilize emulsions, and considering that the rate at which an emulsion breaks down depends on the environmental conditions during its lifetime, the overall goal of this research was to determine the influence of pH and salt concentration (as some of the major factors) on the stability of oil-in-water emulsions.

2. Materials and methods

2.1. Materials

Sunflower oil was obtained from the Institute of Standards and Industrial Research of Tehran; Iranian gum tragacanth was extracted from *A. gossypinus* collected in Isfahan province. The raw gum was ground and the resulting powder was sieved with a mesh of between 200 and 500 microns. The sodium azide (0.02% for sample preservation), HCl and NaCl used in this study were analytical grade, and were purchased from Merck® (Darmstadt, Germany). Double-distilled water was used to prepare all solutions and colloidal dispersions.

2.2. Preparation of gum dispersions

All experiments were performed at room temperature (25 °C). An *A. gossypinus* dispersion (0.5 wt%) consisting of 0.5 g of gum powder (200–500 μm), 99.5 g of distilled water and sodium azide

(0.02 wt%) was stirred for at least 1 h. The solution was stored at 4 °C for about 24 h to ensure complete hydration of the gum.

2.3. Preparation of oil-in-water emulsions

Emulsions were prepared according to Karimi and Mohammadifar (2014). The gum dispersions (90 wt%) and sunflower oil (10 wt%) were homogenized using an Ultraturax (IKA T25, Deutschland, Germany) for 15 min at 13,500 rpm. Emulsions were ice-coated to avoid temperature variation. Salt-stability emulsions (pH 4.0) were prepared with different amounts of NaCl (0, 0.5 and 1 wt%). The emulsions then were stirred thoroughly for 5 min to ensure complete solution of the salt. The pH-stability emulsion samples were prepared using either HCl (0.1 or 1 N) or NaOH (0.1 or 1 N) solution, and the pH was adjusted to the preferred final value (2.5, 4.0 and 5.4).

2.4. pH measurements

Series of 10 wt% oil-in-water emulsions were prepared, and the pH meter (Mettler, Toledo 320) was calibrated with standard solutions of known pH (4.0, 7.0 and 9.0). The pH measurements of the emulsions were read precisely from the instrument and the means of two successive measurements were taken.

2.5. Creaming stability

Thirteen milliliters of each emulsion sample was transferred into a cylindrical test tube (internal diameter 10 mm, height 120 mm), then stored for 150 days at 25 °C. After storage a number of emulsions separated into a thin "creamed" layer at the top and a transparent "serum" layer at the bottom. The height of the initial emulsion (HE) and the height of the serum layer (HS) were determined. The emulsification-stability index (ESI) was calculated as follows (Eq. (1)):

$$(ESI)\% = \frac{(HE - HS) \times 100}{HE} \quad (1)$$

Checking tests were performed in duplicate and the mean of the two single trials was obtained for analysis.

2.6. Zeta-potential

Zeta-potential (z-potential) measurements were determined to provide information about the electrical charge of any droplet formed. Charges were determined (with sample dilution 1:2000) using a Brookhaven Zeta Sizer 90 plus for z-potential at different pH values and salt concentrations. NaCl was added to the water to fix the ionic strength. The pH was adjusted with solutions of HCl and NaOH, and the suspension was conditioned for 1 h before use to measure z-potential. The z-potential measurements are reported as the average of two separate trials.

2.7. Interfacial tension

Interfacial tension (γ) is defined as the energy needed to form a unit of new surface between two immiscible liquids. The interfacial tension between aqueous gum tragacanth solution and sunflower oil was measured according to the Du Nouy ring-pull method using a Krüss digital tensiometer K100 (Krüss Instruments, Germany) equipped with a standard ring probe and an SV 20 glass vessel at 25 °C ± 0.5 °C. A platinum ring was rinsed with distilled water and acetone, and re-rinsed with distilled water, then passed through a blue flame and cooled before each measurement. Twenty grams of the oil phase was carefully layered on top of 30 g of the aqueous phase and left for 30 min at 25 °C before each test. The maximum

force needed to pull the ring from one phase to another is equal to the interfacial tension between the two immiscible liquids. Each sample was analyzed three times; the data are reported as the average. The standard deviation of the interfacial measurements was better than 0.2 mN/m.

2.8. Particle size

The particle size distributions of the non-aggregated emulsions were determined by static laser diffraction using a Cilas 1090 particle-size analyzer (Orleans, France) equipped with a helium–neon laser beam of 5 mW at 635 nm and at ambient temperature (25 °C). Measurements were performed immediately after the emulsion preparation at least in duplicate; and the data are presented as the average. Prior to analysis, each emulsion sample was diluted 1:100 in deionized water (the pH of the samples was adjusted with HCl or NaOH, and ionic strength was changed by adding NaCl). It was important to perform the dilution using water with similar properties to the continuous phase in which the droplets were originally dispersed (that is, having the same salt concentration and pH), and to place the diluted samples immediately into the measurement cell of the device. The samples were then stirred in recirculation water until they achieved an obscuration level of 8–11% as detected by the measurement cell.

Size measurements were reported as volume mean diameter and number mean diameter, as shown in Eqs. (2) and (3), respectively.

$$D[4, 3] = \frac{\sum n_i d_i^4}{\sum n_i d_i^3} \quad (2)$$

$$D[1, 0] = \frac{\sum n_i d_i^1}{\sum n_i} \quad (3)$$

where n_i is the number of particles of class “i”, and d_i is the diameter of class “i”.

Also, the distribution width of droplet size, known as the polydispersity index (span), was calculated using (Eq. (4)):

$$\text{Span} = \frac{D_{0.9} - D_{0.1}}{D_{0.5}} \quad (4)$$

where $d_{0.1}$, $d_{0.5}$ and $d_{0.9}$ are diameters at 10%, 50%, and 90% cumulative volume, respectively (McClements, 2004).

2.9. Rheology measurements

Steady-shear viscosity, strain and frequency-sweep oscillatory shear tests were performed by using a rheometer Anton-Paar (Graz, Austria) MCR 301 model equipped with a concentric cylinder measurement system at room temperature. The temperature was controlled with a Peltier system. Rheological data were collected using Rheoplus software version 3.21 (Anton-Paar). Flow curves were obtained at shear rates of 0.1–1000 s⁻¹. The power law (Eq. (5)) was used to describe rheological properties of emulsions.

$$\eta_a = m \dot{\gamma}^{n-1} \quad (5)$$

where η_a is the apparent viscosity (Pa s), m is the consistency coefficient (Pa sⁿ), $\dot{\gamma}$ is the shear rate (s⁻¹) and n is the flow behavior index (dimensionless).

Strain-sweep tests were performed at a strain of 0.01–600% and a fixed frequency of 1 Hz. To evaluate these values, the critical-strain value limiting the linear viscoelastic region (γ_L), the structural strength (G' at LVE range); the loss modulus at LVE range; and the damping factor ($\tan \delta = G''/G'$) or the ratio of the loss modulus to elastic modulus gave a direct view of whether the samples were liquids or solids, and determined their energy of cohesion (Eq.

(6)) which can be used as a measure of the extent and strength of inter droplet interactions (Tadros, 2004).

$$E_c = \frac{1}{2} G'_{LVE} \times \gamma_{LVE}^2 \quad (6)$$

Dynamic viscoelasticity frequency-sweep tests were carried out at a frequency of 0.1–100 Hz and a constant strain of 1% to calculate the dynamic rheological properties, such as G' and G'' . The Bohlin model ($G' = a\omega^b$) was fitted on the experimental data to determine the structural strength (a) and type of structure (b).

2.10. Statistical analysis

All the experiments in this study were carried out using at least two freshly prepared samples. The statistical analysis was performed by analysis of variance (ANOVA) and test of least significant difference (Duncan's multiple-range; $p < 0.05$) using SPSS version 21. All data were reported as means at $\pm 95\%$ confidence limits.

3. Results and discussion

3.1. Effect of pH and salt concentration on emulsion stability

The plastic test tubes containing the emulsions were visually monitored for gravitational phase separation during 150 days at ambient temperature. The dependence of emulsion stability of creaming on pH and NaCl concentration is shown in Fig. 1. Emulsions that have High emulsion-stability index will generally be more stable against mechanisms of emulsion instability. For the emulsion at pH 2.5, phase separation started after 20 days and the emulsification-stability index decreased gradually with time, reaching 30% after 150 days. Other samples were stable throughout the 150 days, with an emulsification-stability index of 100%. This is due to the fact that electrostatically stabilized emulsions are sensitive to the ionic strength (added salt concentration) and pH of the continuous phase (Hunter, 1998). Although we had initially assumed that the salt concentration influenced the emulsion stability, minerals reduced stability, probably due to an electrostatic screening effect. However, sunflower oil-in-water emulsions prepared with gum tragacanth were stable at all salt concentrations. Therefore the results indicate that salt concentrations do not play a significant role in emulsion stability, and that the effect of pH is greater than that of salt content. Also, it should be observed that gum tragacanth was able to create highly stable emulsions at pH 4.0 after 150 days. This result showed the possibility of using gum tragacanth as an emulsifier for dairy foods and beverages like doogh (Iranian yoghurt drink), as these products also have a pH of 4.0. When the pH started to approach the pKa of gum tragacanth (about pH 3.0), emulsion stability decreased with decreasing pH. This was probably because of the reduction in electrostatic repulsion between droplets; according to the DLVO theory, the energy barrier is responsible for inhibiting droplets from coming near enough to aggregate. Decrease in the electrostatic repulsion decline the height of the energy barrier, until it is not enough to inhibit the droplets from aggregating. Our results agree with those of Farzi et al. (2013), who affirm that gum tragacanth imparts stability to emulsions through electrostatic interactions (that is, a combination of electrostatic and steric repulsion). Also, Jayme, Dunstan, and Gee (1999) similarly reported that neither pH nor calcium chloride concentration has an effect on emulsions stabilized by gum Arabic and modified starch.

3.2. Particle size distribution of emulsions

The droplet size distribution in emulsion can be used to determine the emulsion stability (Huang, Kakuda, & Cui, 2001).

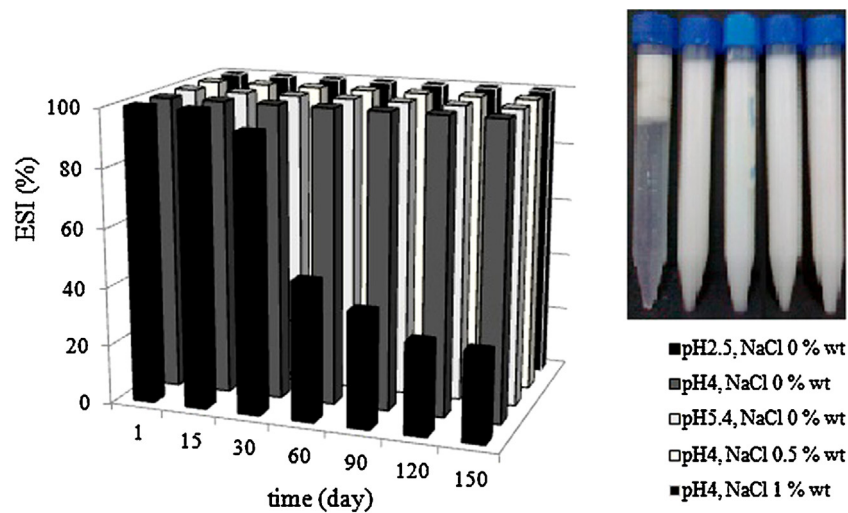


Fig. 1. Photographs of bottle test emulsions and Emulsion stability indexes after 150 days storage at 25 °C for samples containing gum tragacanth (0.5% w/w) at the different pH values and salt concentrations.

Table 1

Particle size analysis of emulsions containing 0.5% w/w gum tragacanth at the different pH values and salt concentrations.

[NaCl] (w/w %)	pH Value	Particle size parameters							
		$D_{0.9}$	$D_{0.5}$	$D_{0.1}$	Span	$D[1,0]$	$D[4,3]$	$D[4,3]A$ (μm)	$D[4,3]B$ (μm)
0	5.4	286.4 ^a	32.82 ^a	3.06 ^b	8.63 ^b	0.81 ^a	91.99 ^a	92.51 ^a	93.41 ^a
0	4.0	234.26 ^b	20.85 ^c	2.57 ^c	9.10 ^a	0.80 ^a	72.94 ^b	73.03 ^b	81.37 ^b
0	2.5	189.57 ^c	25.53 ^b	3.47 ^a	9.29 ^a	0.80 ^a	68.28 ^c	72.64 ^b	–
0	4.0	234.26 ^a	0.80 ^a	2.75 ^b	9.10 ^{ab}	20.85 ^b	72.94 ^b	73.03 ^b	81.37 ^c
0.5	4.0	191.75 ^c	0.81 ^a	2.75 ^b	9.37 ^a	19.55 ^c	53.30 ^c	54.31 ^c	68.76 ^b
1	4.0	228.7 ^b	0.81 ^a	4.73 ^a	8.58 ^b	34.19 ^a	83.71 ^a	83.89 ^a	87.22 ^a

– Phase separation occurred. Values with different letters (a–c) in a column are significantly different ($p < 0.05$). A: $D[4,3]$ on 15th and B: $D[4,3]$ on 150th.

Information about the initial particle size distribution of an emulsion is useful for calculating its long-term stability to creaming, flocculation, coalescence and Ostwald ripening. Particle size parameters including $D_{0.1}$, $D_{0.5}$, $D_{0.9}$, $D[4,3]$, $D[0,1]$ and span values for oil-in-water emulsions stabilized by gum tragacanth depend on the pH and sodium chloride concentration; the values determined in this study are presented in Table 1. Also considering that any increase in the size or particle aggregation can change the volume mean diameter thus this parameter are reported on days 15 and 150 for stable samples. It was found that $D_{0.1}$, $D_{0.5}$, $D_{0.9}$, $D[4,3]$ and span were sensitive to the volume of particles, and that $D[1,0]$ was more sensitive than the other parameters to the number of particles in the distribution. The commonly used method of expressing the mean particle size of a polydisperse emulsion is the volume mean diameter. The results showed that with a decrease in the pH, $D[1,0]$ does not change significantly, and the volume mean diameter ($D[4,3]$) of the droplets decreases from 91 μm to 68 μm . The emulsion at pH 5.4 exhibited the greatest stability and the highest droplet size. This may be explained by the fact that decreasing pH reduces the electrostatic repulsion between droplets, and may also influence the molecular conformation and therefore the molecules' functional properties, by screening electrostatic interactions, binding to negatively charged groups and thus decreasing the number of similarly charged groups. This usually causes particles to become smaller. These results were in good agreement with the conclusions reported by previous studies (Launay, Doublier, & Cuvelier, 1985; Rha & Pradipasena, 1986). Also, the results indicated that the salt concentration altered the emulsion droplet size. Emulsions have a tendency to reduce their volume mean diameter, and $D[1,0]$ showed no significant alterations. It seems that the effect of salt concentration and pH on droplet size have similar reasons. The

increase in volume mean diameter is observed in relatively high concentrations of (1 wt%). This increase in size may be due to change in the conformation of gum tragacanth adsorbed to the surface of emulsion droplets. When the amount of ionic strength increases above a certain level, a salting-out effect occurs. Similar results have been reported by Kulmyrzaev and Schubert (2004) for oil-in-water emulsions stabilized by whey protein isolate. In accordance with Stokes' law, the rate at which droplets move is proportionate to the square of their radius; thus the emulsion stability can be expected to be higher when the droplet size is smaller. However, a correlation was not observed between droplet size distribution and the emulsification-stability index of emulsions during the storage time. The fact that ions influence the interactions between emulsion droplets in so many different ways means that it is difficult to accurately guess or quantify their effect on emulsion properties. Farzi et al. (2013) also found similar behavior in emulsions, with their results indicating that those species of gum tragacanth with the highest volume mean diameter ($D[4,3]$) create the most stable emulsions. As shown in Table 1 the volume mean diameter generally increased over the storage period of 150 days as expected.

3.3. Interfacial tension

Interfacial tension influences emulsion stability by affecting the composition and properties of the interface formed (McClements, 2004). Since correlation was not observed between droplet size distribution and emulsification-stability index, the stability mechanism of emulsion was investigated by interfacial tension measurements. Previous studies have shown that gum tragacanth can decrease the interfacial tension between oil and water, and therefore facilitate emulsion formation (Farzi et al., 2013; Karimi

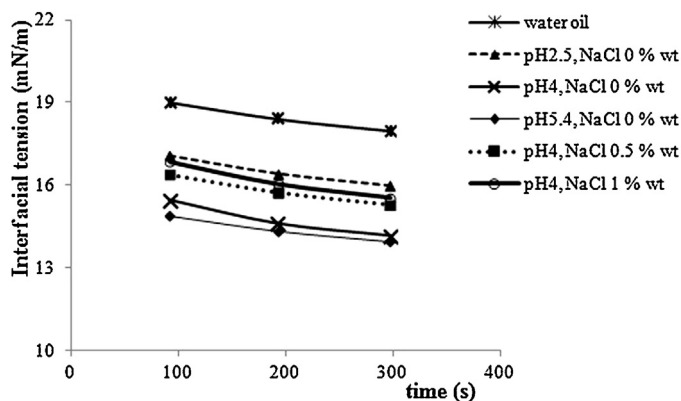


Fig. 2. Interfacial tension as a function of time for emulsion samples containing gum tragacanth (0.5% w/w) at the different pH values and salt concentrations.

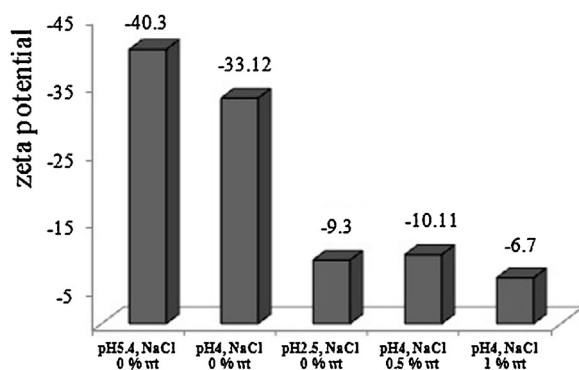


Fig. 3. Effect of pH and salt concentration on Zeta potential of oil in water emulsion samples containing gum tragacanth (0.5% w/w).

& Mohammadifar, 2014). To determine the effect of pH and salt concentration on the emulsifying activity of gum tragacanth, the emulsion samples, including 0.5 wt% gum tragacanth to reduce the interfacial tension of emulsions, were prepared. The dependence of the interfacial tension on pH and salt concentration is shown in Fig. 2. The interfacial tension determined at the sunflower oil–water interface was nearly 18.39 mN/m. The interfacial tension of oil and water was reduced to 14.63 mN/m in the presence of gum tragacanth with no change of pH. There was an increase in interfacial tension with decreasing pH and increasing salt concentration. The interfacial tension of oil and water increased to 14.71, 16.43, 15.76 and 16.04 mN/m for pH 4.0 > 2.5, and salt concentration 0.5 and 1, respectively. Therefore, the results indicated that the pH and salt concentration of the aqueous phase have a strong influence on the interfacial properties of *A. gossypinus*.

It has been proposed that this gum-surface activity is due to the existence of galactose units and protein residue in its structure (Balaghi et al., 2010). Based on the influence of pH and ionic strength on the molecular conformation of gum tragacanth and the number of ionic charges, decreasing pH and increasing salt concentration will increase interfacial tension.

3.4. Zeta-potential

Zeta-potential is one of the tests carried out to determine the behavior of hydrocolloids at the oil–water interface (Garti, Slavin, & Aserin, 1999; Jayme et al., 1999). Fig. 3 shows the zeta-potential alterations of emulsions produced with gum tragacanth at different pH values and salt concentrations. The results show that the maximum zeta-potential value was 40.3 mV for the sample with pH 5.4, and that the zeta-potential decreased with increasing

Table 2

Parameters of power-law model for the emulsion samples containing gum tragacanth (0.5% w/w) at various pH values and salt concentrations.

[NaCl] (w/w %)	pH value	Power law parameters			
		m (Pa s ⁿ)	n	R^2	SD
0	5.4	2.353 ^a	0.473 ^b	0.993	0.832
0	4.0	1.67 ^b	0.481 ^b	0.991	0.834
0	2.5	0.974 ^c	0.539 ^a	0.991	0.634
0	4.0	1.67 ^a	0.481 ^b	0.991	0.834
0.5	4.0	1.195 ^b	0.525 ^a	0.991	0.598
1	4.0	1.019 ^c	0.521 ^a	0.992	0.505

SD: Standard Deviation, R^2 : The coefficient of determination. Values with different letters (a–c) in a column are significantly different ($p < 0.05$).

salt concentration and with decreasing pH. This trend is expected, since an increase in salt concentration and decrease in pH will lead to a decrease of the electrostatic repulsion and a corresponding reduction in zeta-potential. These results are in agreement with previous measurements of the destabilization of emulsions (Acedo-Carrillo et al., 2006; Jayme et al., 1999). In the current study, the emulsion-stability tests showed that sunflower oil-in-water emulsions prepared with gum tragacanth were stable at all salt concentrations after 150 days. However, in this study the zeta-potential by itself could not indicate as successfully as the other factors whether the emulsion would be stable. Hence, more information about the mechanism of stabilization by gum tragacanth was necessary; thus, this study measured the steady- and unsteady-state rheological properties of the samples.

3.5. Rheological properties

3.5.1. Steady-shear tests

A rheological study of an emulsion is an important factor to consider, as it can provide useful information on an emulsion's stability and inner microstructure. The correlation between the rheological properties of oil-in-water emulsions and the physicochemical characteristics of the continuous phase has been investigated (McClements, 2004; Tadros, 2004). All samples exhibited non-Newtonian behavior (shear-thinning), in that the apparent viscosity decreased with increasing the shear rate. As the shear rate increases, flocs are broken down to smaller sizes and the emulsion viscosity decreases. It has been shown that gums exhibit a non-Newtonian rheology. Also, this behavior agrees with previously available data for gum tragacanth (Balaghi et al., 2010; Dagleish, 2003). Among the emulsion samples, pH 5.4 produced the highest viscosity, up to almost 0.694 Pa s at the lowest shear rate of 10 s^{-1} . One of the main factors in emulsion stability is high viscosity, which inhibits the coalescence of the oil droplets (Dickinson, Radford, & Golding, 2003). With decreasing pH and increasing salt concentration, the apparent viscosity decreased for all samples. The Power-law model was applied to explain the shear-thinning behavior of emulsions. As shown in Table 2, with decreasing pH and increasing salt concentration, the consistency coefficient (m) decreased and the flow-behavior index (n) increased. The consistency coefficient for samples with different pH followed the order pH 5.4 > pH 4.0 > pH 2.5, and that for samples with different salt concentrations followed the order 0% > 0.5% > 1%. Ions have been found to decrease viscosity of hydrocolloids and their emulsions because of compaction of the electrical double layer and the decrease in the effective molecular volume of the gum (Tadros, 2013; Weiping, 2000); this is in agreement with decreases in the emulsion-stability index and droplet size.

3.5.2. Oscillatory measurements

The rheological parameters of strain sweep test are shown in Table 3 as a function of pH and salt concentration. It was found

Table 3

Structural strength (G'_{LVE}) and G''_{LVE} , cohesive energy (E_c), limiting value of strain (γ_{LVE}) and loss-tangent value ($\tan \delta_{LVE}$) in the linear viscoelastic range of emulsions containing gum tragacanth (0.5% w/w) at various pH values and salt concentrations, as determined by amplitude sweep tests at 25 °C and a frequency of 1 Hz.

[NaCl] (w/w%)	pH value	G'_{LVE} (Pa)	G''_{LVE} (Pa)	γ_{LVE}	E_c (J/m ³)	$\tan \delta_{LVE}$
0	5.4	4.65 ^a	5.77 ^a	6.99 ^c	114 ^a	1.26 ^c
0	4.0	3.19 ^b	4.52 ^b	8.17 ^b	106 ^b	1.42 ^b
0	2.5	1.21 ^c	2.32 ^c	12.6 ^a	96 ^c	1.92 ^a
0	4.0	3.19 ^a	4.52 ^a	8.17 ^b	106 ^b	1.42 ^c
0.5	4.0	1.37 ^c	2.41 ^b	7.97 ^c	43.5 ^c	1.76 ^a
1	4.0	1.75 ^b	2.34 ^c	11.3 ^a	111 ^a	1.61 ^b

Values with different letters (a–c) in a column are significantly different ($p < 0.05$).

that emulsions with pH 5.4 had the highest dynamic modulus (an indicator of structural strength) at the linear viscoelastic range. The emulsions studied demonstrated a large linear viscoelastic range, which depends on pH values and salt concentrations. Emulsions at pH 2.5 had the highest value for γ_{LVE} (limiting value of reversible strain-induced structural change), at 12.6. Furthermore, cohesive energy (which indicates the extent and strength of inter-droplet interactions) decreased with decreasing pH and increasing salt concentration. However, an increase in cohesive energy was observed at relatively high salt concentrations (1 wt%); this agrees with the data for G'_{LVE} and droplet size. An examination of the relationship between rheological properties and pH and salt concentration showed that the rheological properties of electrostatically stabilized emulsions with small droplets were mainly sensitive to pH and salt concentration. The storage shear modulus usually initially decreases with increasing salt concentration because of the resulting screening of the charges. However, past a particular salt concentration, the interactions between the emulsion droplets become attractive more than repulsive, and thus flocculation will take place, causing an increase in storage shear modulus. The rheological properties of electrostatically stabilized systems are consequently particularly sensitive to both pH and salt concentration (Keowmaneechal & McClements, 2002; McClements, 2004; Weiss & McClements, 2000). The ratio of G'' to G' ($\tan \delta$) reported in Table 3 provided a direct analysis for whether the samples acted as liquids or solids. For all samples, this parameter was greater than 0.1, which revealed the liquid-like feature of emulsions (Farzi et al., 2013; Krstonosic, Dokic, Dokic, & Dapcevic, 2009). Viscoelastic properties (storage modulus (G') and loss modulus (G'')) for emulsions stabilized by 0.5 wt% gum tragacanth at different pH values and salt concentrations are shown in Fig. 4A and B, respectively, as a

Table 4

Bohlin model parameters for the elastic modulus of 10% w/w oil in water emulsions containing 0.5% w/w gum tragacanth at various pH values and salt concentrations as determined by frequency sweep tests at 25 °C and a strain of 1%.

Bohlin model parameters				
[NaCl] (w/w%)	pH value	a	b	R^2
0	5.4	1.63 ^{6a}	0.590 ^c	0.998
0	4.0	0.874 ^b	0.725 ^b	0.998
0	2.5	0.261 ^c	0.814 ^a	0.995
0	4.0	0.874 ^a	0.725 ^b	0.998
0.5	4.0	0.285 ^c	0.737 ^a	0.857
1	4.0	0.335 ^b	0.741 ^a	0.991

Values with different letters (a–c) in a column are significantly different ($p < 0.05$).

function of frequency. For samples with pH 5.4 and pH 4.0 and samples with salt concentrations of 0, 0.5 and 1 there was a change from a predominantly viscous response at a lower frequency ($G'' > G'$) to a predominantly elastic response at a higher frequency ($G' > G''$). The crossover point took place at the frequency of 15.82 for pH 5.4 and 6.31 Hz for emulsions pH 4.0. For samples with different salt concentrations the crossover occurred at the frequency of 6.31 for 0%, 2.51 for 0.5% and 4.09 for 1%.

The crossover point presents good information about the molecular weight of ingredients, the extent and strength of physical and chemical bonds and the viscoelastic behavior of materials. The lower crossover point showed that the contribution of viscosity was larger (Farzi et al., 2013). No crossover value was observed for samples with pH 2.5.

This behavior indicated a weakening of emulsions' microstructure when the pH came close to the pKa of gum tragacanth. Samples with pH 5.4 emulsion were more solid-like than others. A similar pattern emerged in samples based on their salt concentration. The frequency dependence of the storage modulus (G') for emulsion samples might be described by the Bohlin model. Related data are summarized in Table 4. Significant differences were found in the a -values and b -values for the different emulsion samples. By decreasing pH and increasing salt concentration, a -values, an indicator of structural strength, decreased drastically and b -values, an indicator of the type of structure (which is the slope of G' versus ω plot), increased. Samples with pH 2.5 had the lowest a -value and the highest b -value, and were unstable. Samples with a salt concentration of 1% showed an increase in structural strength (a -value). These results confirmed the conclusion exhibited in the previous sections. Therefore the low stability of pH 2.5 was complemented

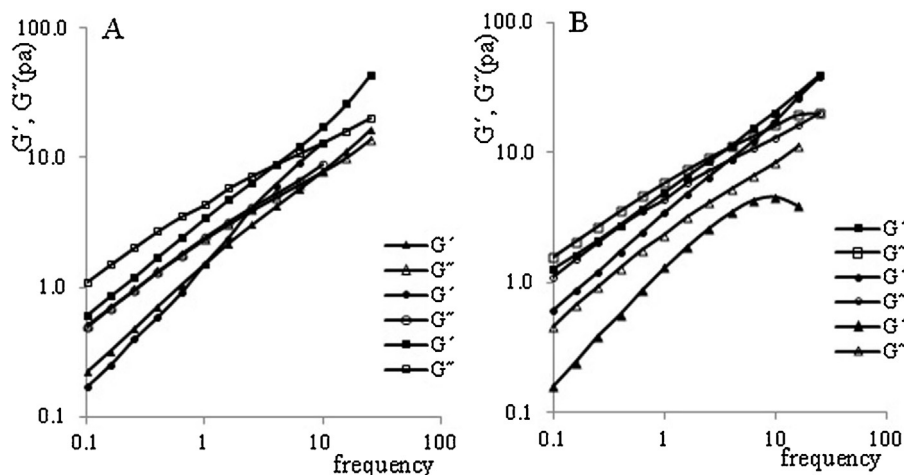


Fig. 4. Frequency sweeps of modulus G' and G'' for 10% w/w oil in water emulsions containing 0.5% w/w gum tragacanth (25 °C) (A) at the pH values ■ 5.4, ● 4, ▲ 2.5, (B) at the salt concentrations ■ 0, ● 0.5%, ▲ 1%.

by the lowest values for G'_{LVE} , cohesive energy and a -value, and the highest values for $\tan \delta$ and b -value.

4. Conclusions

According to the results, it can be concluded that emulsions stabilized with gum tragacanth show resistance to instability mechanisms when exposed to a range of salt concentrations (0–1 wt% NaCl) after 150 days of storage at room temperature. Also, this study has shown that the stability of emulsions is strongly dependent on pH. Emulsion droplets were extremely unstable to aggregation near the pKa of gum tragacanth because of the low electrostatic repulsion between the droplets. Confirmatory and complementary relationships were found between rheological and particle-size parameters. Our results have significant consequences for the potential use of gum tragacanth in food emulsions.

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