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Emulsifying properties of water soluble yellow mustard mucilage: A comparative study with gum Arabic and citrus pectin

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ABSTRACT

The emulsifying properties of water soluble yellow mustard mucilage (WSM) were compared with two commercial emulsifiers—gum Arabic and citrus pectin. Of the three polysaccharides examined, WSM exhibited excellent emulsion stability under various conditions applied in this study. The emulsion stability test showed that 1% and 2% WSM emulsions exhibited no phase separation after 21 days of storage at 25 °C. WSM also exhibited the highest surface activity among the three materials. The surface activity of 1% WSM is equivalent to that of a 15% gum Arabic solution. When exposed to heat at 90 °C for 20 min, WSM emulsion showed little change in droplet size, which was superior to both the gum Arabic and pectin emulsions, whose droplet size increased significantly following heat treatment. WSM emulsion also had a significantly larger magnitude of zeta-potential than the other two polysaccharides. Under different pH conditions, droplet size varied greatly among the three materials. WSM emulsions exhibited significantly smaller droplet size at pH 4.0 followed by pH 9.0 and 6.5. Pectin emulsions and gum Arabic emulsions were not sensitive to the pH range examined in this study. WSM also showed the best freeze–thaw stability among the three polysaccharides. After three freeze–thaw cycles, the 0.5% WSM emulsion exhibited a higher cream index than 2% gum Arabic emulsion or the 1% pectin emulsion.

1. Introduction

Yellow mustard mucilage, the dried layer on yellow or white mustard seeds (*Sinapis alba*, L.), is readily extracted by water at room or elevated temperature. The yield of the crude extract was 5% of the total seed weight with the water-soluble portion accounting for 55.6% of the crude extract (Cui, Eskin, Wu, & Ding, 2006). Our previous studies have shown that the soluble fraction was composed of 84% pectic polysaccharides and 16% non-pectic polysaccharides, the pectic fraction of which exhibits shear-thinning flow behaviour while the non-pectic fraction forms gels when exposed to heat, acid and alkaline. The water-soluble mucilage also interacts synergistically with galactomannans (Cui et al., 2006; Wu, Cui, Eskin, & Goff, 2009, 2011). Previous studies have examined the chemical, structural, and rheological properties of yellow mustard mucilage and its various fractions, however, the emulsifying properties of the water-soluble fraction have not been well documented. Gum Arabic is a naturally-occurring complex polysaccharide with a small amount of protein (2.5%) integrated into the highly branched polysaccharide consisting of galactose, arabinose, rhamnose and glucuronic acid (Williams & Phillips, 2009). Its emulsifying properties appear to be attributed to hydrophobic protein chains which adsorb onto the surface of oil droplets while hydrophilic polysaccharide fractions limit oil droplet aggregation and coalescence by steric and/or repulsive electrostatic forces (Dickinson, 2009).

At present, the food industry has placed a significant amount of attention on pectins for their role as stabilizers in oil-in-water emulsions (Leroux, Langendorf, Schick, Vaishnav, & Mazoyer, 2003). They consist of α-1,4 linked galacuronic acid chains with occasional 1,2 linked L-rhamnose, neutral sugar residues linked to rhamnose as branches. It contains some carboxyl units which provide more repulsive forces among molecules, thus increasing the stability of the emulsion system (Williams & Phillips, 2009). Leroux et al. (2003) demonstrated that citrus pectin and beet pectin can efficiently reduce the interfacial tension between the oil and

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water phases in the emulsions, however, their emulsion stabilizing properties vary due to differences in acetyl groups, proteins and calcium ion.

The objective of this study was to examine the emulsifying properties of WSM in oil-in-water emulsions in comparison to gum Arabic and citrus pectin. In addition to measuring the interfacial activities of the gum solutions, we also compared emulsion stability, oil distribution, zeta-potential and freeze–thaw stability at different pH and temperature conditions. The results of this study are intended to provide technical information to the food industry supporting the potential of WSM as a novel ingredient for food emulsions.

2. Materials and methods

2.1. Materials

Gum Arabic (from acacia tree, G-9752, synonyms *Acacia senegal* gum), and citrus pectin (from citras peel, P-9135, with ≥74.0% galacturonic acid and ≥6.7% methoxy group) were purchased from Sigma-Aldrich (Saint Louis, USA). WSM was prepared according to the method described by Wu et al. (2009). Vegetable oil (pure soybean oil) was purchased from a local grocery store (Great Value Brand, Wal-Mart Stores, Inc., Bentonville, AR, USA).

2.2. Solution preparation for surface tension measurement

Each sample solution was prepared with 5 concentrations for surface activity measurements. For gum Arabic, the concentrations used were 15.00, 10.00, 5.00, 1.00 and 0.5% (w/w), respectively. For pectin and WSM, the concentrations used were 1.50, 1.00, 0.50, 0.25 and 0.10% (w/w), respectively. Gum Arabic solutions were prepared by dissolving the sample in distilled water at room temperature with constant stirring for 12 h before measurements. Pectin and WSM were prepared by dissolving the samples in distilled water at 70 °C for 1 h with constant stirring followed by cooling at room temperature with constant stirring for 2 h before measurements.

2.3. Preparation of oil-in-water emulsion

Aqueous solutions of gum Arabic, pectin and WSM were prepared in 5 mM phosphate buffer at pH 6.5. Gum Arabic solution was prepared at room temperature for 12 h with constant stirring, while pectin and WSM solutions were prepared at 70 °C for 1 h and followed by cooling at room temperature for 2 h with constant stirring. For pH studies the samples were dissolved in 5 mM phosphate buffer with pH values adjusted to 4.0, 6.5 and 9.0 respectively using NaOH or HCl. Aqueous solutions were mixed with 10% (w/w) vegetable oil and pre-homogenized using a polytron (PT 2100, Kinematica AG, Schweiz) for 2 min, then passed 3 times through a high pressure homogenizer (Microfluidizer LV1, Microfluidics Inc., Newton, MA) at 11,000 psi.

2.4. Methods

2.4.1. Emulsion stability

Emulsion stability was measured over 21 days at room temperature (21 °C) by comparing the evolution of phase separation. The thickness of the serum layer (Cao, Dickinson, & Wedlock, 1990) and creamed phase (gradient creaming, Laplante, Turgeon, Paquin, 2002, 2004) were measured as percentages of total emulsion height in tubes (34 mm). The measurement was recorded at day 21.

2.4.2. Surface tension

Surface tension of air–water interface was measured according to the method described by Izydorczyk, Biliaderis, and Bushuk (1991). The decrease in surface tension (mN/m) of water with increased concentration of samples was measured by the Du Nouy ring method using a tensiometer (Attension Sigma 702, Biolin Scientific, USA). The force acting on the ring was measured as it was moved upward from an air-gum dispersion surface. Equal volumes of solution (50 ml) were placed in 100 ml glass beakers. The changes in surface tension were recorded every 10 min for 2 h at 22.0 ± 0.5 °C. The value of the surface tension was the average of the 12 measurements from 10 min to 120 min.

2.4.3. Zeta-potential and droplet size

Zeta-potential and droplet size (d.nm) were determined using Zetasizer 90 (Malvern Instruments Ltd, USA). All measurements were carried out according to the method described by Gu, Decker, and McClements (2007) with slight modifications. For zeta-potential measurement, emulsions were diluted to 0.05% w/w droplet concentration using 5 mM phosphate buffer at pH 6.5. Diluted emulsions were injected into a high-concentration vial specifically designed for zeta-potential. For measurement of droplet size, the emulsion samples were diluted to approximately 0.005% w/w using 5 mM phosphate buffer (pH 6.5). The measurement of particle size was performed using a Dynamic Light Scattering instrument which measures the rate of fluctuation in scattering intensity and then uses this to calculate the size of the particles. Mie theory was used to convert intensity size distribution to volume distribution. The analysis was performed at a scattering angle of 90° at a temperature of 22 °C. Mean droplet size (d.nm) was used to represent the size distribution. Gu et al. (2007) pointed out that dilution was necessary to avoid multiple scattering effects, even though this step might disrupt the emulsion system. Since this was applied to all samples, the results were comparable to one another.

2.4.4. Influence of pH and temperature

The effect of pH was examined by preparing a series of emulsion samples at pH 4.0, 6.5 and 9.0 for zeta-potential and particle size measurement. For the effect of temperature, emulsion samples were incubated at 90 °C for 20 min and then cooled down to room temperature by placing the tubes in ice water. After treatments, samples were stored for 3 h and then tested for droplet size and zeta-potential analysis.

2.4.5. Freeze–thaw treatment

The influence of freeze–thaw cycle on emulsion was determined using the method described by Gu et al. (2007). Emulsion samples (10 ml) were all incubated in a −20 °C freezer for 22 h. After incubation the samples were thawed for 2 h at 40 °C. This freeze–thaw cycle was repeated 3 times with the creaming stability compared among the emulsion samples.

2.4.6. Statistical analysis

Three polysaccharide types – WSM, gum Arabic and pectin – were evaluated under three concentration levels (0.5, 1.0 and 2.0% w/w) or three pH levels (4.0, 6.5 and 9.0). A balanced design with three replicates for each of the nine combinations of treatments was set up. As per the experimental design, fresh samples were prepared, measured three times and checked for any discrepancy in measurement. Those three measurements were then averaged to estimate a measurement for each sample. Each sample is independent and prepared randomly. Two-way ANOVA with Tukey's HSD multiple comparison tests were performed in R statistical computing environment (R Development Core Team, 2014). *P*